Aspirin Desensitization Achieved After Omalizumab Treatment in a Patient With Aspirin-Exacerbated Urticaria and Respiratory Disease

Guillén D[1], Bobolea I[1], Calderon O,[1] Fiandor A,[1] Cabañas R[1], Heredia R[1], Quirce S[1,2]

[1]*Department of Allergy, Hospital La Paz Institute for Health Research (IdiPAZ), Madrid, Spain*

[2]*CIBER de Enfermedades Respiratorias CIBERES, Madrid, Spain*

Key words: Aspirin-exacerbated respiratory disease (AERD). Aspirin desensitization. Non-steroidal anti-inflammatory drugs (NSAIDs). Omalizumab. Aspirin-exacerbated urticaria.

Palabras clave: Enfermedad respiratoria exacerbada por aspirina (EREA). Desensibilización a aspirina. Anti-inflamatorios no esteroideos (AINE). Omalizumab. Urticaria exacerbada por aspirina.

Aspirin-exacerbated respiratory disease (AERD), formerly known as aspirin and nonsteroidal anti-inflammatory drug (NSAID) intolerance or idiosyncrasy, is characterized by the concomitant presence of nonallergic hypersensitivity to NSAIDs and a respiratory disease of variable clinical expression, namely chronic rhinosinusitis (with or without nasal polyposis) and bronchial asthma, which is exacerbated (naso-ocular reaction and/or bronchoconstriction) by therapeutic or diagnostic exposure to a NSAID [1,2]. AERD management should be multidisciplinary, and comprise, on one hand, medical and surgical treatment of the various underlying diseases and their possible complications, including desensitization in selected cases, and on the other hand, avoidance of NSAIDs with provision of effective therapeutic alternatives [3,4]. Avoidance of NSAIDs does not ensure disappearance of the airway inflammation; on the contrary, asthma and polyposis usually continue to progress and persist for life. Aspirin desensitization is indicated in patients with uncontrolled bronchial and nasal symptoms or multiple polypectomies, in patients who require long-term treatment with oral corticosteroids, and in patients with AERD who need NSAID treatment for specific diseases, such as rheumatic or cardiovascular conditions [5]. Desensitization followed by long-term daily doses of aspirin improves upper and lower respiratory symptoms and sense of smell, decreases the number of emergency visits and hospitalizations for asthma, increases quality of life associated with rhinitis and asthma perception, and reduces the need for new polypectomies and for topical and systemic corticosteroids [5,6]. AERD patients often have co-existing conditions, such as urticaria or respiratory allergies, and it is therefore important for clinicians to recognize these additional triggers and treat them in order to optimize the overall management of the disease [4].

We report on a 48-year old woman with a 20-year history of chronic rhinosinusitis with nasal polyposis requiring multiple sinus operations in addition to asthma and chronic idiopathic urticaria. She had experienced worsening of nasal congestion, rhinorrhea, wheezing, and urticaria on several occasions following NSAID intake (ibuprofen 600 mg, aspirin 500 mg). She thereafter avoided these drugs, as advised, and experienced no more episodes of urticaria. When first seen at our department in 2011, she reported good tolerance to paracetamol 650 mg but not to paracetamol 1 g, which triggered the above-mentioned reactions. Her asthma symptoms were controlled with salmeterol/fluticasone 50/250 mcg twice a day. However, because of severe nasal symptoms consisting of congestion, hyposmia, and frequent sinus infections (score of 35 on a 100-mm visual analog scale for nasal symptoms) and regrowth of nasal polyps, she was waiting for her third endoscopic sinus operation. She denied seasonal worsening of respiratory symptoms. Skin prick tests with common aeroallergens were negative, and baseline spirometric parameters were normal (forced vital capacity [FVC], 128% of predicted; forced expiratory volume in the first second [FEV_1], 100.8%; FEV_1/FVC, 77.35). A bronchodilator test was positive (increase in FEV_1 of 14% and 420 mL). A specific bronchial challenge with lysine acetylsalicylate was negative, so we carried out an oral challenge with aspirin [3]. Two hours after the administration of 50 mg of aspirin the patient developed pruritic urticaria on her face and body. There was no dyspnea and the pulmonary auscultation was normal. The cutaneous symptoms subsided 24 hours after the intravenous administration of corticosteroids and antihistamines. The patient reported that this is how her symptoms typically started, with progression to upper and lower respiratory involvement, when she had taken full doses of NSAIDs in the past. A subsequent oral challenge with etoricoxib (cumulative dose, 90 mg) was also positive, with generalized hives but no respiratory symptoms. The patient was diagnosed with NSAID hypersensitivity with cutaneous and respiratory involvement (aspirin-exacerbated urticaria and respiratory disease).

The patient underwent endoscopic sinus surgery for the third time in the ear, nose, and throat (ENT) department, without complications; she reported persistence of anosmia at discharge. To decrease the risk of nasal polyp recurrence and improve overall symptom control and quality of life, aspirin desensitization was requested by both the ENT specialist and the patient, in spite of our reticence regarding possible outcomes in view of the presence of associated urticaria. Having provided signed, informed consent, the patient received pretreatment with montelukast 10 mg/d and continued to take salmeterol/fluticasone 50/250 mcg twice a day. Two hours after initiation of the oral aspirin desensitization protocol, with 25 mg of aspirin, the patient developed pruritic urticarial papules on her face, neck, arms, and back (Figure A). Spirometric parameters remained normal.

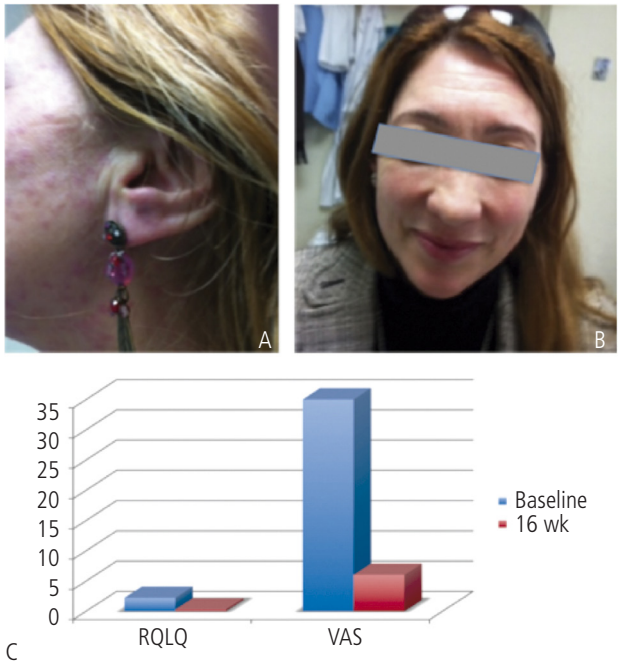

Figure. Tolerance of aspirin desensitization before and after omalizumab treatment. A, First desensitization, 2 hours after the administration of 25 mg aspirin. B, Second desensitization: tolerance of 650 mg aspirin after omalizumab treatment. C, RQLQ and VAS scores at baseline and 16 wk. RQLQ indicates Rhinoconjunctivitis Quality of Life Questionnaire; VAS, visual analog scale.

Initial improvement of the cutaneous lesions was observed after intravenous antihistamines and corticosteroids, but 1 hour later the cutaneous lesions on her face worsened, with the additional development of palpebral angioedema. New lesions also appeared on her legs. Complete remission was not achieved until 1 week later after daily treatment with levocetirizine and a short course of oral corticosteroids. Aspirin desensitization was consequently interrupted.

Off-label treatment with omalizumab was initiated at a dose of 150 mg every 4 weeks (weight 68 kg, total IgE, 108 kU/L) for 16 weeks, with a dual purpose: to prevent regrowth of the nasal polyps [7] and to reattempt aspirin desensitization, based on the hypothesis that omalizumab would be able to prevent the urticaria [8]. A new desensitization protocol, with the same conditions as above, was started after 16 weeks of treatment with omalizumab. The patient achieved tolerance of 650 mg of aspirin after 10 days of desensitization consisting of a more gradual protocol than usual [5] (Figure B, C).

Now, almost 2 years later, the patient tolerates 300 mg twice daily of aspirin and requires omalizumab 150 mg/mo. She is completely free of both respiratory and cutaneous symptoms. She does not require asthma control treatment, has not experienced any nasal polyp recurrences to date, and has a slightly improved sense of smell. Moreover, on several occasions she has tolerated ibuprofen 600 mg 3 times daily without developing any cutaneous symptoms. We attempted to withdraw omalizumab on 2 occasions (after 6 and 12 months of treatment), but the urticaria returned 5 to 6 weeks after the last dose of omalizumab.

We have reported on a case of AERD in which aspirin desensitization was indicated [3,9]; the case was complicated by the coexistence of NSAID-induced urticaria and angioedema. This association is quite common in clinical practice, since both diseases share physiopathological mechanisms consisting of a series of chronic alterations to the arachidonic acid metabolic pathways. So, unlike chronic urticaria, aspirin- and NSAID-induced urticaria is explained by the dose-dependent effects of cyclooxygenase 1 inhibition after NSAID administration [2,3,10].

To our knowledge, this is the first case of aspirin-induced urticaria successfully treated with omalizumab. The use of omalizumab is hereby justified, as it was instrumental in achieving—and maintaining—aspirin desensitization. Omalizumab therapy was requested by the patient as it is the only treatment to date that has proven capable of modifying the natural course of AERD [6].

Funding

The authors declare that no funding was received for the present study.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

References

1. Quiralte J, Blanco C, Castillo R, Delgado J, Carrillo T. Intolerance to non-steroidal antiinflammatory drugs: results of controlled drug challenges in 98 patients. J Allergy Clin Immunol. 1996;98:678-85.
2. Berges-Gimeno MP, Simon RA, Stevenson DD. The natural history and clinical characteristics of aspirin-exacerbated respiratory disease. Ann Allergy Asthma Immunol. 2002;89:474-8.
3. Kowalski ML, Makowska JS, Blanca M, Bavbek S, Bochenek G, Bousquet J, Celik G, Demoly P, Gomes ER, Nizankowska-Mogilnicka E, Romano A, Sanchez-Borges M, Sanz M, Torres MJ, De Weck A, Szczeklil A, Brockow K. Hypersensitivity to nonsteroidal anti-inflammatory drugs (NSAIDs) – classification, diagnosis and management: review of the EAACI/ENDA and GA2LEN/HANNA. Allergy. 2011;66:818-29.
4. Lee RU, Stevenson DD. Aspirin-exacerbated respiratory disease: evaluation and management. Allergy Asthma Immunol Res. 2011;3:3-10.
5. Stevenson DD. Aspirin desensitization in patients with AERD. Clin Rev Allergy Immunol. 2003;24:159-68.
6. Berges-Gimeno MP, Simon RA, Stevenson DD. Long-term treatment with aspirin desensitization in asthmatic patients with aspirin-exacerbated respiratory disease. J Allergy Clin Immunol. 2003;111:180-6.
7. Vennera Mdel C, Picado C, Mullol J, Alobid I, Bernal-Sprekelsen M. Efficacy of omalizumab in the treatment of nasal polyps. Thorax. 2011;66:824-5.
8. Kaplan AP, Joseph K, Maykut RJ, Geba GP, Zeldin RK. Treatment of chronic autoinmune urticaria with omalizumab. J Allergy Clin Immunol. 2008;122:569-73.

9. Alobid I, Antón E, Armengot M, Chao J, Colás C, del Cuvillo A, Dávila I, Dordal MT, Escobar C, Fernández-Parra B, Gras-Cabrerizo JR, Ibáñez MD, Lluch M, Matéu V, Montoro J, Gili JR, Mullol J, Navarro AM, Pumarola F, Rondón C, Sánchez-Hernández MC, Sarandeses A, Soler R, Valero AL. SEAIC-SEORL Consensus Document of Nasal Polyposis - POLINA Project. J Investig Allergol Clin Immunol. 2011;21(Suppl 11):1-58.
10. Stevenson DD, Sanchez-Borges M, Szczeklik A. Classification of allergic and pseudoallergic reactions to drugs that inhibit cyclooxygenase enzymes. Ann Allergy Asthma Immunol. 2001;87:177-80.

Manuscript received December 30, 2013; accepted for publication, May 19, 2014.

Daiana Guillén
Departamento de Alergología
Hospital Universitario La Paz (IdiPAZ)
Paseo de la Castellana, 261
28046 Madrid, Spain
E-mail: dguillen27@gmail.com

Herpes-like Eruption Due to Fluconazole

González-Fernández T[1], López-Freire S[1], Juangorena M[1], Méndez-Brea P[1], Vázquez-Veiga H[2]
[1]Allergy Department, Complejo Hospitalario Universitario de Santiago, Santiago de Compostela, Spain
[2]Dermatology Department, Complejo Hospitalario Universitario de Santiago, Santiago de Compostela, Spain

Key words: Fluconazole. Fixed-drug eruption. Herpes-like.

Palabras clave: Fluconazol. Erupción fija medicamentosa. Tipo herpes.

Oral azole drugs, such as ketoconazole, fluconazole, and itraconazole, among others, represent a major advance in antifungal therapy. Hypersensitivity reactions to these drugs are uncommon [1], despite the high frequency of interactions. The case reported herein describes an atypical and infrequent manifestation of a hypersensitivity drug reaction to fluconazole.

A 30-year-old woman with no personal or family history of allergy was referred to our allergy department complaining of a skin reaction she had experienced on repeated occasions over the past 3 months. She had been previously diagnosed with cutaneous psoriasis but had not needed treatment in the preceding 2 years. On her first visit, she described the skin lesions as papules with vesicles and crusts on the lower lip with patchy areas of facial erythema. The first episode had occurred 2 hours after having Christmas dinner and she reported contact with a new colored cream that she had applied 8 hours beforehand. She had experienced a second and a third episode 2 and 3 months later. She reported having eaten nuts and chocolate ice-cream 2 hours before the reactions, which consisted of recurrence of lesions at the same location and the appearance of similar vesicular rashes on the elbows and a small rash on the abdomen. Once totally free of lesions she was specifically asked about, and denied, any contact with drugs.

An allergological study including skin prick tests with a range of food extracts (including nuts, shellfish, egg, milk, fresh fruits, legumes, pork and beef, profilin, lipid transfer protein from peach, gliadin, fish, and spices) was negative in all cases. Patch tests with the European Standard Series and the patient's cosmetic products were performed and read at 48 and 96 hours. These tests were also all negative. Due to the implication of ice-cream, an ice cube test was carried out, but no positive response was observed. Twenty-four hours after eating the same ice-cream that had been reportedly implicated in the third reaction, the patient developed a localized vesicular rash on her lower lip, followed by new blisters on the elbows and a patchy erythematous rash on her face (Figure). The lesions were diagnosed as a herpes-like rash. However, scraping of the ulcer base in search of giant multinuclear cells (Tzanck cells), which are typical of herpes virus infection, was negative. We did not detect any serological data indicative of a specific viral infection. Again, due to the peculiarity of this clinical case, we insisted on possible contact with a hidden drug or sensitizer.

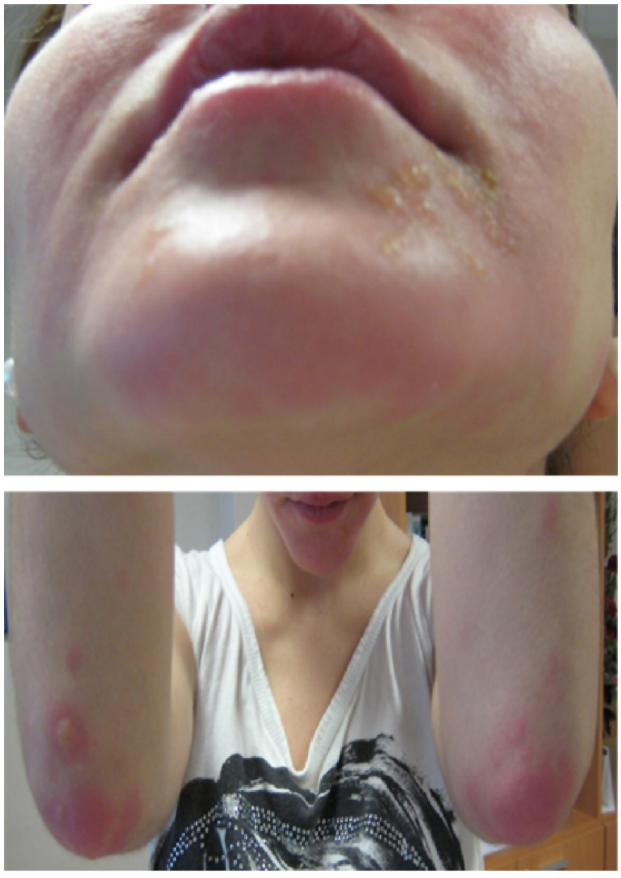

Figure. Vesicles on the lower lip, blisters on the elbows and a patchy erythematous rash on the face.

The patient finally admitted to taking monthly fluconazole to treat recurrent vaginitis. She reported that she had taken oral fluconazole 150 mg 24 hours before each episode. To conclusively prove the relationship between fluconazole and the rash, we performed patch tests with fluconazole 2%, ketoconazole 1%, itraconazole 2%, and voriconazole 2%, which gave negative results at 48 and 96 hours. Intradermal tests with fluconazole 0.002 and 0.02 mg/mL were also negative at 30 minutes and 24 hours. Twelve hours after an oral challenge with fluconazole (cumulative dose of 150 mg), the patient developed itchy, patchy erythema on her face, followed, 24 hours later, by confluent blisters on the lower and upper lip and on both elbows. The patient did not agree to a biopsy. An oral challenge test with itraconazole was performed 1 month later, with no reaction.

To the best of our knowledge, this is the third reported case of a fluconazole herpes-like reaction [2,3], although several cases of fixed drug eruptions due to fluconazole have been reported [4-8]. Some authors consider herpes-like rashes to be a form of fixed drug reaction. Nevertheless, the classic presentation, which consists of single or multiple sharply demarcated nummular plaques leading to hyperpigmentation on the skin, was not present in our patient. Intermittent drug administration is more likely than continuous administration to induce sensitization, and fluconazole is usually prescribed on a monthly basis to treat fungal vaginitis. Patch tests do not seem to be useful in this setting and it would appear that challenge tests are needed to prove the implication of a suspected drug. A thorough, accurate clinical record is essential for detecting the etiological agent, especially in cases of intermittent use.

Funding

The authors declare that no funding was received for this study.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

References

1. Nakai N, Katoh N. Fixed drug eruption caused by fluconazole: A case report and mini-review of the literature. Allergology International. 2013;62:139-141.
2. Benedix F, Schilling M, Schaller M, Röcken M, Biedermann T. A young woman with recurrent vesicles on the lower lip: fixed drug eruption mimicking herpes simplex. Acta Derm Venereol. 2008;88:491-4.
3. Jensen ZN, Bygum A, Damkier P. Fluconazole-induced fixed drug eruption imitating herpes labialis with erythema multiforme. EJD. 2012;22:693-4.
4. Gaiser CA, Sabatino D. Fluconazole-induced fixed drug eruption. J Clin Aesthet Dermatol. 2013;6:44-5.
5. Kim CY, Kim JG, Oh CW. Fluconazole induced fixed drug eruption. Ann Dermatol. 2011;23:S1-3.
6. Morgan JM, Carmichael AJ. Fixed drug eruption with fluconazole. Br Med J. 1994;12:454.
7. Hekkila H, Timonen K, Stubb S. Fixed drug eruption due to fluconazole. J Am Acad Dermatol. 2000;42:883-4.
8. Goel A, Jain C. Fluconazole induced fixed drug eruption a rare offender. J Dermatol. 2004;31:345-6.

Manuscript received February 14, 2014; accepted for publication, May 20, 2014.

Teresa González-Fernández
Servicio de Alergia
Complejo Hospitalario Universitario
de Santiago (Hospital de Conxo)
Rúa Ramón Baltar sn
15706 Santiago de Compostela
Spain
E-mail: teregf8@hotmail.com

Rubinstein-Taybi Syndrome Associated With Humoral Immunodeficiency

Pasic S
Pediatric Immunology, Mother and Child Health Institute, Medical Faculty, University of Belgrade, Serbia

Key words: Rubinstein-Taybi syndrome. Immunodeficiency. Dandy-Walker anomaly.

Palabras clave: Síndrome Rubinstein-Taybi. Inmunodeficiencia. Anomalía Dandy-Walker.

Rubinstein-Taybi syndrome (RTS) is characterized by short stature, mental retardation, broad thumbs and first toes, cardiac abnormalities, feeding difficulties, and recurrent upper respiratory tract infections [1,2].

We report on an 8-year-old girl with RTS. She was first admitted to our hospital at 18 months of age because of hypotonia, developmental delay, and recurrent infections. She is the first child of healthy, nonconsanguineous parents. Her past medical history revealed that she had experienced repeated episodes of otitis media and pneumonia in infancy.

Clinical examination revealed facial features suggestive of RTS, with arched eyebrows, eye skinfolds, and a prominent beaked nose with widely spaced eyes (Figure A, informed consent obtained). She also had pale skin and broad, angulated first toes with partial syndactyly of the second and third toe of the right foot. Chest auscultation revealed bilateral wheezing and normal heart sounds. Neurologic examination revealed generalized hypotonia with the inability to sit or stand without support. A cranial computed tomography scan showed Dandy-Walker anomaly.

Laboratory investigations at admission were as follows: erythrocyte sedimentation rate, 18 mm/h; hemoglobin, 115 g/L, white blood cell count, $10.3x10^9$/L with 52% neutrophils, 38% lymphocytes, and 8% monocytes. The platelet count was $368x10^9$/L. The bone marrow aspirate examination was normal. Immunologic investigations showed very low serum IgA (0.11g/L), IgM (0.13 g/L), and IgG (1.3 g/L) concentrations; serum IgE was undetectable. The patient had received routine vaccines in infancy, but specific antibody responses to tetanus toxoid, polio, diphtheria, and hemophilus influenzae type B were absent. The peripheral blood lymphocyte phenotype was normal for the patient's age. Further analyses performed at 3 years of age revealed decreased numbers of nonswitched IgD^+CD27^+ B cells and switched memory IgD^-CD27^+ B cells (Figure B). The karyotype of peripheral blood lymphocytes revealed 16p chromosome deletion. Fluorescence in situ hybridization confirmed a deletion in the 16p13.3 region.

Occurrence of respiratory infections had previously been explained by microaspirations due to significant gastroesophageal reflux, a feature present in the majority of patients with RTS [2]. Only a few reports have evaluated the immune system in RTS [3]. Villella et al [4] described the case of a 4-year-old patient with RTS who presented with recurrent infections and normal serum IgG levels, but absence of a specific immune response after immunization against pneumococcus. Rivas et al [5], in turn, described a patient with RTS who had defective phagocytosis, decreased T-cell counts, and normal serum immunoglobulins, while Kimura et al [6] described hypoplastic thymus on autopsy of a patient with RTS [6]. In our patient, specific antibody responses to both protein and polysaccharide antigens were absent. Low percentages of both nonswitched and memory B cells were also detected, although this finding may represent a normal variation within the patient's age group [7].

Dandy-Walker-like anomaly associated with humoral immunodeficiency and congenital heart and facial defects has been described as Ritscher-Schinzel syndrome [8]. There have also been rare reports, like ours, of Dandy-Walker anomaly in association with RTS [9,10].

Our patient was given regular intravenous immunoglobulin (IVIG) replacement therapy, which led to a decrease in the frequency of respiratory infections. At the ages of 4 and 8 years, transient hypogammaglobulinemia of infancy was excluded on detection of decreased serum IgG concentrations (<1g/L) when IVIG replacement therapy was temporarily stopped. The patient's serum IgA and IgM concentrations (0.11 g/L and IgM, 0.10 g/L, respectively) remained barely detectable.

The finding of humoral immunodeficiency in our patient may explain the early onset of pyogenic infections. A careful immunologic investigation is required in patients with RTS who present with recurrent infections or unexplained episodes of fever.

Acknowledgments

I would like to thank M. Recher and L.D. Notarangelo at the Children's Hospital in Boston, USA for their phenotypic analysis of B lymphocytes.

Funding

This manuscript is funded by grants (No. 175065 and No. 175073) awarded to SP by the Ministry of Science and Technology, Republic of Serbia.

Conflicts of Interest

The author declares that he has no conflicts of interest.

References

1. Rubinstein JH, Taybi H. Broad thumbs and toes and facial abnormalities. Am J Dis Child 1963;105:588-607.
2. Wiley S, Swayne S, Rubinstein JH, Lanphear NE, Stevens CA. Rubinstein-Taybi syndrome medical guidelines. Am J Med Genet 2003;119A:101-10.
3. Ming JE, Stiehm ER, Graham Jr JM. Immunodeficiency as a component of recognizable syndromes. Am J Med Genet 1996;66: 378-98.
4. Villella A, Bialostocky D, Lori E, Meyerson H, Hostoffer R. Rubinstein-Taybi syndrome with humoral and cellular defects: a case report. Arch Dis Child 2000;83:360-61.

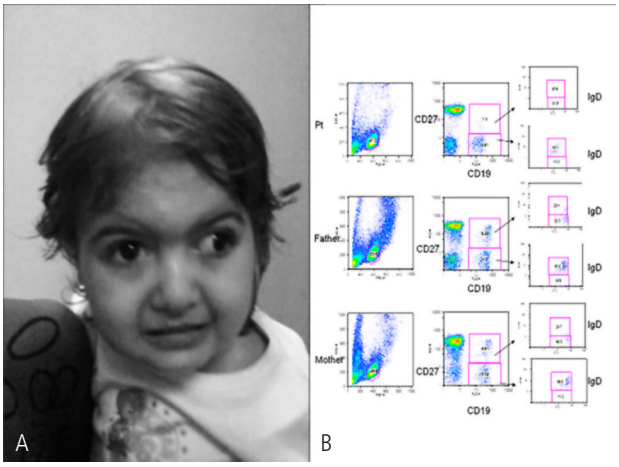

Figure. A, Facial characteristics in our patient: widely spaced eyes, broad nasal bridge, and arched eyebrows. B, Flow-cytometric analysis showing decreased nonswitched B cells and switched memory B cells.

5. Rivas F, Fragoso R, Ramos-Zepeda R, Vaca G, Hernandez A, Gonzales-Quiroga G, Olivares N, Cantu JM. Deficient cell immunity and mild intermittent hyperaminoacidemia in a patient with the Rubinstein-Taybi syndrome. Acta Paediatr Scand 1980;69:123-6.
6. Kimura H, Ito Y, Koda Y, Hase Y. Rubinstein-Taybi syndrome with thymic hypoplasia. Am J Med Genet 1993; 46:293-6.
7. aan de Kerk DJ, Jansen MH, ten Berge IJM, van Leuween EMM, Kuijpers TW. Identification of B cell defects using age-defined reference ranges for in vivo and in vitro B cell differentiation. J Immunol 2013;190:5012-9.
8. Launer R, Seger R, Jorg W,Halle F, Aeppli R, Schinzel A. Immunodeficiency associated with Dandy-Walker-like malformation, congenital heart disease, and craniofacial abnormalities. Am J Med Genet 1989;33:280-1.
9. Bonioli E, Bellini Di Stefano A. Unusual association: Dandy-Walker-like malformation in the Rubinstein-Taybi syndrome. Am J Med Genet 1989;33:420-1.
10. Agarwal R, Aggarwal R, Kabra M, Deorari AK. Dandy-Walker malformation in Rubinstein-Taybi syndrome: a rare association. Clin Dysmorphol 2002;11:223-4

Manuscript received April 5, 2014; accepted for publication, May 20, 2014.

Srdjan Pasic
Mother & Child Health Institute
8 R. Dakica Street
11070 Belgrade
Serbia
E-mail: pasics@ikomline.net

Relief of Photoallergy: Atorvastatin Replacing Simvastatin

Sommer M, Trautmann A, Stoevesandt J
Department of Dermatology and Allergy, University Hospital Würzburg, Germany

Key words: Allergy. Atorvastatin. Photosensitivity. Phototesting. Simvastatin.

Palabras clave: Alergia. Atorvastatina. Fotosensibilidad. Fotoparche. Simvastatina.

The differential diagnosis of skin eruptions confined to air- and UV-exposed areas should include airborne contact dermatitis, autoimmune disorders, idiopathic photodermatoses, and phototoxic/photoallergic reactions to topically applied or systemic photosensitizers. Once a diagnosis of drug-induced photoallergy is suspected, accurate identification and adequate replacement of the elicitor constitute major clinical challenges.

A 71-year-old white man with no history of pre-existing skin disease was started on a number of drugs to treat ischemic heart disease following coronary stent implantation. Medication with bisoprolol, amlodipine, quinapril, hydrochlorothiazide, acetylsalicylic acid, ezetimibe, and simvastatin was well tolerated during the first months of treatment. The patient then took up outdoor cycling in early springtime to further improve his physical fitness. He subsequently developed an itchy rash extending from the head and neck to the dorsal forearms. His condition progressively worsened despite application of topical steroids, necessitating inpatient treatment by mid June.

On admission to hospital, the patient had a pruritic eczematous rash on UV-exposed skin of the dorsal hands, neck, head, and face, which was accompanied by pronounced periorbital edema; the rash spared the submental area (Figure A-C). Routine laboratory examinations including a full blood count, C-reactive protein, liver and renal function tests, serum cholesterol, and triglycerides revealed normal findings. Serum total IgE was within the normal range (21.0 kU/L). Antinuclear antibodies with a speckled staining pattern (titer 1:320) were detected on HEp-2 cells. There was, however, no evidence of autoantibodies to extractable nuclear antigens (including Ro/SSA, La/SSB, and Jo-1) or anti-dsDNA. Direct immunofluorescence did not reveal any specific deposits, and histology revealed spongiotic eczematous dermatitis.

Due to suspicion of drug-induced photoallergy, treatment with hydrochlorothiazide was stopped. The skin eruptions, however, returned almost immediately after the patient was discharged from hospital, and slowly subsided only after subsequent discontinuation of simvastatin.

Allergologic and photodiagnostic work-up was initiated 2 months after complete resolution of the skin lesions. Patch tests and photopatch tests including standard contact allergen series, hydrochlorothiazide, and simvastatin did not yield any positive reactions. The minimal erythema dose (MED) was

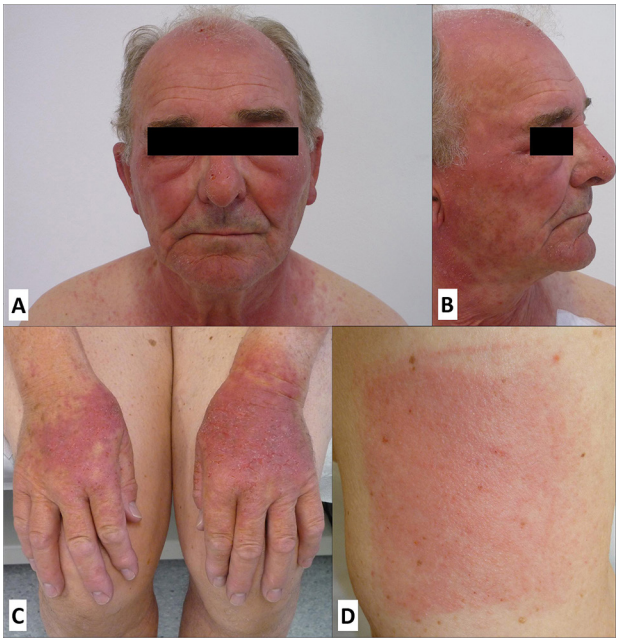

Figure. A-C, Eczematous dermatitis on sun-exposed skin of the dorsal hands, neck, head, and face accompanied by pronounced periorbital edema, but sparing the submental area. D, UV-A photoprovocation testing following 10-day intake of 40 mg simvastatin: eczematous reaction with marked epidermal thickening, papules, and small vesicles.

determined with sources of broadband UV-B and UV-A. UVB-MED (75 mJ/cm-^{2}) and UVA-MED (30 J/cm-^{2}) were within the normal range. Photoprovocation testing with broadband UV-A (10 J/cm-^{2}) and broadband UV-B (75 mJ cm-^{2}) was carried out by irradiating an area of 5×8 cm on the patient's dorsal upper arm on 2 successive days. Readings were recorded 24, 48, and 72 hours postirradiation, with results showing mild solar erythema. Photoprovocation testing was repeated 10 days after restarting simvastatin at a daily dose of 40 mg. Again, only mild solar erythema was observed in the UV-B-irradiated area. A progressive erythematous reaction with marked epidermal thickening, papules, and small vesicles, however, developed in UV-A-irradiated skin, reaching its maximum 72 hours postirradiation (Figure D). Histology confirmed a spongiotic eczematous reaction pattern.

A definitive diagnosis of simvastatin-induced photoallergy with an action spectrum in the UV-A range was made, and our patient was advised to permanently avoid simvastatin. A routine medical check-up some 12 months later revealed a rise in cholesterol levels and progression of ischemic heart disease, prompting the treating cardiologist to enquire whether he might prescribe an alternative statin to replace simvastatin. We agreed on treatment with atorvastatin, which was well tolerated. Twelve months later, our patient is still taking atorvastatin. He has remained free of skin symptoms while safely enjoying different kinds of outdoor activities.

Drug-induced photoallergy is a delayed-type hypersensitivity reaction, which clinically and histologically resembles allergic contact dermatitis. Radiation, most commonly in the UV-A range, is essential to form a complete photoallergen [1]. Allergologic work-up is complex and necessitates a stepwise approach beginning with selective discontinuation of the most likely elicitor to achieve clinical stabilization prior to testing. Simultaneous discontinuation of multiple drugs should be avoided as this impedes identification of the culprit photosensitizer, while exposing the patient to an increased medical risk. Photopatch testing of drugs is poorly standardized and may yield false negative results, in particular if hepatic metabolism of the causative drug is required for antigen formation. In this case, repeated photoprovocation testing with and without systemic intake of the putative culprit drug remains the ultimate confirmatory test.

Statins are not commonly considered to belong to the top group of photosensitizing substances, which comprises a number of antibiotics (eg, fluoroquinolones, tetracyclines, sulphonamides), nonsteroidal anti-inflammatory analgesics (eg, naproxen, piroxicam), psychoactive drugs (eg, phenothiazine), amiodarone, and thiazide diuretics [2]. Statins have been marketed to lower cholesterol levels by inhibition of the enzyme 3-hydroxy-3-methylglutaryl (HMG) coenzyme A reductase since the early 1980s. International treatment guidelines strongly advocate their use for secondary prevention in early stages of ischemic heart disease [3]. Systemic adverse effects are rare, but may be serious, as in the case of statin-induced rhabdomyolysis. Case reports have documented a variety of cutaneous statin-induced adverse effects that may be attributed to the immunomodulatory properties of this group of drugs. These adverse effects include subacute cutaneous lupus erythematosus [4], dermatomyositis [5], lichenoid drug eruptions [6], and photoallergic reactions [7-9]. Photoallergic reactions may take a chronic course despite withdrawal of the statin [8] or present as noneczematous erythema multiforme [9]. Photodermatitis is most frequently attributed to simvastatin, which is the most widely prescribed statin drug. Little is known about the safety of other statins in pre-existing simvastatin-induced photoallergy. We based our decision to support treatment with atorvastatin on a comparison of molecular structures. All statins share an HMG-like moiety that occupies the enzyme active site of HMG coenzyme A reductase [10]. Simvastatin, lovastatin, and pravastatin (also referred to as type 1 statins) are structurally similar, sharing a common decalin ring and a butyryl substituent. Fully synthetic type 2 statins (fluvastatin, rosuvastatin, and atorvastatin) have a common fluorophenyl group and different ring structures linked to the HMG-like moiety [10]. We hypothesize that a switch from type 1 to type 2 statins (and possibly vice versa) represents a safe therapeutic option in statin-induced photoallergy.

Funding

The authors declare that no funding was received for the present study.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

References

1. González E, González S. Drug photosensitivity, idiopathic photodermatoses, and sunscreens. J Am Acad Dermatol. 1996;35(6):871-885.

2. Ferguson J. Photosensitivity due to drugs. Photodermatol Photoimmunol Photomed. 2002;18(5):262-269.
3. Stone NJ, Robinson J, Lichtenstein AH, Merz CN, Blum CB, Eckel RH, Goldberg AC, Gordon D, Levy D, Lloyd-Jones DM, McBride P, Schwartz JS, Shero ST, Smith SC Jr, Watson K, Wilson PW. 2013 ACC/AHA Guideline on the Treatment of Blood Cholesterol to Reduce Atherosclerotic Cardiovascular Risk in Adults: A Report of the American College of Cardiology/ American Heart Association Task Force on Practice Guidelines. Circulation. 2013. [Epub ahead of print].
4. Suchak R, Benson K, Swale V. Statin-induced Ro/SSa-positive subacute cutaneous lupus erythematosus. Clin Exp Dermatol. 2007 Sep;32(5):589-91.
5. Inhoff O, Peitsch WK, Paredes BE, Goerdt S, Goebeler M. Simvastatin-induced amyopathic dermatomyositis. Br J Dermatol. 2009;161(1):206-208.
6. Roger D, Rolle F, Labrousse F, Brosset A, Bonnetblanc JM. Simvastatin-induced lichenoid drug eruption. Clin Exp Dermatol. 1994;19(1):88-89.
7. Morimoto K, Kawada A, Hiruma M, Ishibashi A, Banba H. Photosensitivity to simvastatin with an unusual response to photopatch and photo tests. Contact Dermatitis. 1995;33(4):274.
8. Granados MT, de la Torre C, Cruces MJ, Piñeiro G. Chronic actinic dermatitis due to simvastatin. Contact Dermatitis. 1998;38(5):294-5.
9. Rodríguez-Pazos L, Sánchez-Aguilar D, Rodríguez-Granados MT, Pereiro-Ferreirós MM, Toribio J. Erythema multiforme photoinduced by statins. Photodermatol Photoimmunol Photomed. 2010;26(4):216-218.
10. Istvan ES, Deisenhofer J. Structural mechanism for statin inhibition of HMG-CoA reductase. Science. 2001;292(5519):1160-1164

Manuscript received February 7 2014; accepted for publication, May 26, 2014.

Johanna Stoevesandt
Department of Dermatology and Allergy
University Hospital Würzburg
Josef-Schneider-Straße 2
D - 97080 Würzburg
Germany
E-mail: Stoevesandt_J@ukw.de

Allergic Reaction to Undeclared Lupin in a Chocolate

Eguíluz Gracia I[1], Martínez González de Lema B[1], Rubio-Pérez M[1], Ruíz-Giménez L[1], Recio Blázquez L[2], Pastor-Vargas C[3], Fernández-Rivas M[1]

[1]*Allergy Department, Hospital Clínico San Carlos, IdISSC, Madrid, Spain*

[2]*Hospital Pharmacy Department, Hospital Clínico San Carlos, Madrid, Spain*

[3]*Immunology Department, IIS-Fundación Jiménez Díaz, Madrid, Spain*

Key words: Lupin allergy. Hidden allergen. Undeclared allergen.

Palabras clave: Alergia a lupino. Alérgenos ocultos. Alérgenos no declarados.

Although lupin (*Lupinus albus*) has been consumed as a snack for many years, it has only recently been introduced as a cereal substitute by the food industry. Its growing use has been accompanied by reports of allergic reactions, including respiratory symptoms after lupin inhalation and local or generalized reactions following ingestion [1]. Attempts to determine population threshold doses for lupin that elicit allergic reactions have been unsuccessful due to considerable interpatient variability [2]. Because of these difficulties and increasing reports of allergic reactions to lupin, the 2006 European Commission Directive included lupin in its mandatory labeling list, whereby lupin must always be listed as a food ingredient, irrespective of the amount present [3].

We report the case of a 30-year-old atopic woman who developed an itchy throat, cough, and shortness of breath shortly after eating a pepper and lemon chocolate. The symptoms disappeared in 2 hours with an oral antihistamine. The patient had never experienced oral pruritus after the ingestion of any food. One week before the reported reaction she developed mild urticaria affecting the arms and legs that subsided within 24 hours. She could not relate this reaction to the ingestion of any specific foods. The skin prick test was positive for cat dander (mean wheal diameter, 8 mm), dog dander (4 mm), grass pollen (6.5 mm), lupin (15 mm) and soybean (3.5 mm), and negative for milk, celery, egg, mustard, sesame, wheat, *Anisakis simplex*, latex, peach, tomato, tree nuts, peanut, legumes, mites, molds, and weed and tree pollens (Laboratorios Leti). The prick-prick test was positive for lupin (10 mm) and the pepper and lemon chocolate (5 mm). The serological study (ImmunoCAP, ThermoFisher Scientific Phadia) showed specific IgE (sIgE) for lupin (42.2 kU_A/L), chickpea (3.12 kU_A/L), vetch (0.90 kU_A/L), and carob (0.68 kU_A/L). sIgE levels were under 0.35 kUa/L for celery, sesame, pepper, Pru p 3, tree nuts, peanut, and other legumes. Total IgE and baseline tryptase levels were 85.9 and 2.22 kU_A/L, respectively.

The patient had eaten the pepper and lemon chocolate from a box of assorted chocolates. The labeled ingredients were cocoa, soy lecithin, milk, egg, sugar, sorbitol, honey, lemon essence, cayenne pepper, and unspecified flour. The

manufacturer denied the use of lupin in both the pepper and lemon chocolate and other foods processed nearby. Between the time of the reaction and her visit to our department, the patient had followed a normal diet and tolerated chocolate, lemon and other fruits, peanut, soybean, lentils, and sunflower seed. An open oral challenge excluded clinical reactivity to chickpea.

Lupin and the culprit chocolate were extracted as previously described [4] and SDS-PAGE was carried out under reducing conditions. Polyacrylamide concentrations of 14% (wt/vol) and 5% (wt/vol) were used for the separating and stacking gels, respectively. Twenty micrograms of protein extract was applied per lane and protein electrophoresis was performed for each extract. The separated proteins were transferred to nitrocellulose membranes for immunoblot analysis according to the method described by Benito et al [5]. The blocked membranes were washed and cut into strips for separate incubation with untreated patient serum, or serum previously incubated with either lupin or chocolate as previously described [6]. The strips were then washed and incubated with anti-human IgE antibody conjugated with horseradish peroxidase (SouthernBiotech). Finally, the presence of IgE-binding bands was visualized by enhanced chemiluminescence (GE Healthcare) following the instructions provided by the manufacturer. Serum binding to proteins exhibited a similar pattern in both extracts (Figure, A,B, lane 1). Serum preincubation with none of the extracts was able to inhibit the recognition of bands in both the lupin and chocolate extracts (Figure, lanes 4 and 5). Serum preincubation with bovine serum albumin did not affect band recognition in the lupin extract (Figure, lane 3), and serum from a negative control individual was not able to bind proteins in the lupin extract (Figure, lane 2).

Since the patient had experienced the reaction after the ingestion of an "unconventional" chocolate, and had previously developed mild self-limited urticaria, we decided to investigate clinical reactivity to lupin and explore its potential severity by means of a double-blind placebo-controlled food challenge (DBPCFC). Because of the risk of a reaction after the ingestion of, for instance, a lupin-containing spicy food that could induce confusing oral symptoms, we decided to skip the oropharyngeal mucosa by administering encapsulated lupin flour. This could trigger severe reactions, but also provides important information for risk management decisions. The patient was fully informed and provided written consent. The DBPCFC was performed by trained staff, with full equipment and medication readily available. An intravenous line was inserted. Lactose-filled capsules were prepared as placebo and increasing amounts of lupin flour were introduced into identical capsules for the up-dosing challenge protocol (0.5, 1, 3, 10, 30, 100 and 300 mg). The patient tolerated the placebo but developed epigastralgia, generalized urticaria, and conjunctivitis 20 minutes after the ingestion of the 300-mg lupin capsule (cumulative dose of 444.5 mg). The symptoms disappeared within 3 hours of the administration of intramuscular epinephrine plus intravenous antihistamines and corticosteroids. A significant increase in serum tryptase was observed, from 3.7 kU_A/L at the beginning of the reaction to 7.41 kU_A/L at 60 minutes and 16.0 kU_A/L at 120 minutes. The diagnosis of lupin allergy was established and a lupin-free diet was recommended. The patient was advised to read all food labels carefully and to carry rescue medication including self-injectable epinephrine. At the time of writing, the patient is still on a lupin-free diet and has had no further reactions.

According to the chocolate box label, flour was one of the ingredients. The use of lupin was denied by the manufacturer, without any further specifications. As the immunological study revealed full cross-reactivity of the patient's serum with both lupin and chocolate extracts, we think that this unspecified flour was lupin flour. The dose that elicited the reaction during the challenge was within the range previously reported for lupin [7]. In this case, lupin behaved as a hidden allergen [8]. This report reveals that despite current regulation, it appears that there are still manufacturers that do not report the presence of lupin as an ingredient and also emphasizes the need for adequate control of food production, manipulation, and labeling processes.

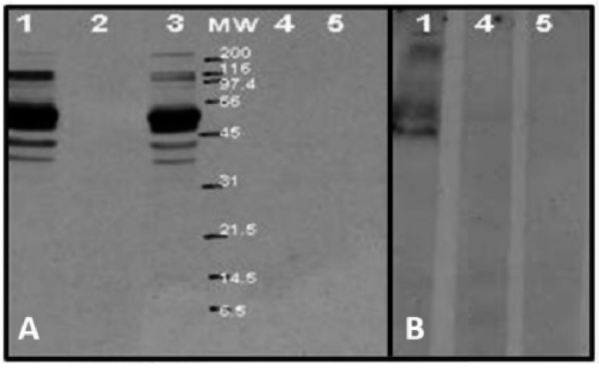

Figure. Immunoblot analysis of lupin (panel A) and chocolate (panel B) extracts. Lanes 1, Patient's serum (noninhibited). Lane 2, Negative control. Lane 3, Patient's serum inhibited with BSA. MW, Molecular weight markers (kDa). Lanes 4, Patient's serum inhibited with lupin extract. Lanes 5, Patient's serum inhibited with chocolate extract.

Funding

The authors declare that no funding was received for the present study.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

References

1. Campbell CP, Yates DH. Lupin allergy: a hidden killer, a menace at work; occupational disease due to lupin allergy. Clin Exp Allergy. 2010;40(10):1467-72.
2. Jappe U, Vieths S. Lupine, a source of new as well as hidden food allergens. Mol Nutr Food Res. 2010;54:113-26.
3. European Commission. Commission Directive 2006/142/EC of 22 December 2006 amending Annex IIIa of Directive 2000/13/EC of the European Parliament and of the Council listing the ingredients which must under all circumstances appear on the labeling of foodstuffs. Off J Eur Union. 2006; L 368:110-1.

4. Guillamon E, Rodriguez J, Burbano C, Muzquiz M, Pedrosa MM, Cabanillas B, Crespo JF, Sancho AI, Mills EN, Cuadrado C. Characterization of lupin major allergens (Lupinus albus L.). Mol Nutr Food Res. 2010;54:1668-76.
5. Benito C, González-Mancebo E, Alonso-Díaz de Durana D, Tolón RM, Fernández-Rivas M. Identification of a 7S globulin as a novel coconut allergen. Ann Allergy Asthma Immunol. 2007;98:580–4.
6. Pérez-Gordo M, Cuesta-Herranz J, Maroto AS, Cases B, Ibáñez MD, Vivanco F, Pastor-Vargas C. Identification of sole parvalbumin as a major allergen: study of cross-reactivity between parvalbumins in a Spanish fish-allergic population. Clin Exp Allergy. 2011;41(5):750-8.
7. NDA (Scientific Panel on Dietetic Products, Nutrition and Allergies). Opinion of the scientific panel on dietetic, products, nutrition and allergies on a request from the commission related to the evaluation of lupin labelling purposes. The EFSA Journal 2005; 302:1-11.
8. Sanz ML, de Las Marinas MD, Fernandez J, Gamboa PM. Lupin allergy: a hidden killer in the home. Clin Exp Allergy. 2010;40(10):1461-6

Manuscript received April 1, 2014; accepted for publication, May 26, 2014.

Montserrat Fernández Rivas
Hospital Clínico San Carlos
Servicio de Alergia
c/ Prof. Martín Lagos s/n
28040 Madrid, Spain
E-mail mariamontserrat.fernandez@salud.madrid.org

Anaphylaxis in a Child After Ingestion of Persimmon

Rodríguez-Jiménez B[1], Núñez Acevedo B[1], Ledesma A[2], Cava Sumner B[1], Kindelan-Recarte C[1], Domínguez-Ortega J[3]
[1]*Allergy Unit, Hospital Universitario de Getafe, Madrid, Spain*
[2]*ALK-Abelló, Madrid, Spain*
[3]*Allergy Service, Hospital Universitario La Paz, Madrid, Spain*

Key words: Food allergy. Lipid transfer protein. Persimmon. Sharon fruit.

Palabras clave: Alergia alimentaria. Proteína de transferencia de lípidos. Caqui. Saroni.

Persimmon is a tropical fruit belonging to the Ebenaceae family. It is thought to have antioxidant properties owing to its high content in flavonoids and vitamins A, C, and E. The different varieties of persimmon are classified according to whether they are astringent or not. The nonastringent variety includes Sharon fruit (*Diospyros kaki*). Allergy to this fruit is extremely rare.

We present the case of an 8-year-old boy who was referred to our clinic in December 2011 because he had experienced pruritus, generalized itching, urticaria, labial and palpebral edema, dyspnea, and wheeze while eating Sharon fruit. He had not developed gastrointestinal symptoms or hypotension. He required emergency treatment (inhaled salbutamol, intramuscular adrenaline, dexchlorpheniramine, and intravenous prednisone), which led to resolution of symptoms.

Until then he had eaten persimmon without problems and tolerated banana, avocado, kiwi, chestnut, and peach, as well as other fruits and nuts. He also tolerated contact with latex.

The patient had had rhinoconjunctivitis due to pollen sensitization since the age of 3 years that was being treated with oral antihistamines on demand. He had never received pollen-specific immunotherapy and was not exposed to animals at home.

Skin prick testing was performed with commercial extracts of the most common local pollens, profilin, standardized peach lipid transfer protein (LTP) (ALK-Abelló), fruits, nuts (Leti), and latex. The results were positive for grasses, cypress, plane tree, *Plantago, Artemisia, Chenopodium*, cat dander, standardized peach LTP, avocado, and chestnut. Prick-prick testing with persimmon was positive with both the peel (10 mm) and the flesh (22 mm).

Total IgE was 517 kU_A/L. Specific IgE testing (sIgE) (CAP System, Phadia Thermo Fisher) was performed with the following allergens: plane tree (5.30 kU_A/L), avocado (2.52 kU_A/L), kiwi (3.91 kU_A/L), chestnut (10.00 kU_A/L), and latex (0.48 kU_A/L). sIgE results for recombinant allergens of *Phleum pratense* were as follows: rPhl p 1, 20.70 kU_A/L and rPhl p 5, rPhl p 7 (polcalcin), and Phl p 12 (*Phleum pratense* profilin), <0.35 kU_A/L. sIgE for peach LTP (Pru p 3) was 53.40 kU_A/L.

A persimmon extract was obtained in order to investigate the allergens recognized by the patient. The peel was separated from the flesh and each sample was lyophilized separately.

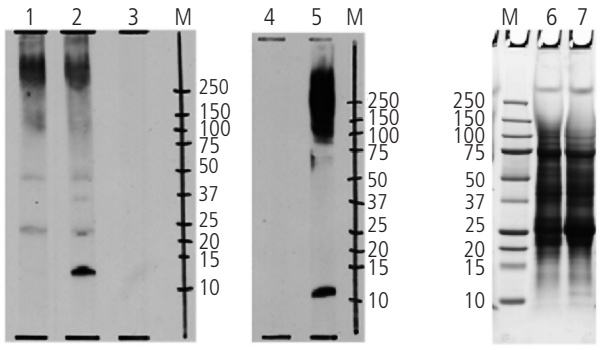

Figure. Lane 1, Flesh extract with patient's serum. Lane 2, Peel extract with patient's serum. Lane 3, Peel + flesh extract (negative control without patient's serum). Lane 4, Peel extract (negative control without polyclonal antiserum). Lane 5, Peel extract incubated with polyclonal anti-LTP rabbit serum. Lane M, Molecular weight markers (KDa). Lane 6, Flesh extract stained with Coomassie blue. Lane 7, Peel extract stained with Coomassie blue.

The allergens were extracted in sodium chloride 1.8% for 90 minutes at 4ºC with magnetic stirring. They were then centrifuged, and the supernatants filtered (0.2 µm).

The peel and flesh extracts and the molecular weight markers were analyzed using Tricine SDS-PAGE under nonreducing conditions (Figure).The proteins separated in the polyacrylamide gel were electronically transferred to strips of nitrocellulose paper [1]. The nitrocellulose strips were saturated with 1% casein in phosphate-buffered saline (PBS) and incubated with the patient's serum diluted at 1:5. As a negative control, a nitrocellulose strip containing the same extract was incubated with 1% casein in PBS. After washing, the strips were incubated with ascitic fluid containing monoclonal antihuman IgE (HE-2) diluted 1:3000 [2]. After additional washing, the strips were incubated with rabbit antimouse antibody conjugated with horseradish peroxidase (RAM-HRP, DAKO) diluted 1:5000. Finally, the strips were washed and the IgE-binding proteins were detected using chemoluminescence (ECL, GE Healthcare). The total quantity of protein in the gel was 60 µg/line.

IgE-reactive bands were observed in both the peel and the flesh extracts. Bands of lower intensity (molecular weights of approximately 22 and 45 kDa) were present in both extracts. However, a band of approximately 12 kDa was observed in the peel only (Figure).

Immunoblotting inhibition was performed. Peel extract was transferred to the nitrocellulose strips, and the patient's serum was added to one of the strips. Serum that had previously been incubated with purified rPru p 3 (5 µg) was added to another strip. (The allergen was provided by Dr Araceli Díaz-Perales's group at the Center for Plant Biotechnology and Genomics (UPM-INIA) in Pozuelo de Alarcon, Madrid, Spain.) No reduction was observed in the signal of the 12-kDa band; in other words, Pru p 3 was unable to inhibit binding of the patient's serum to the persimmon band.

We then performed immunodetection with polyclonal antiserum. The peel extract and molecular weight markers were transferred to nitrocellulose membranes as described above. Once transfer was complete, the strips were saturated in 1% casein in PBS and incubated with a specific polyclonal rabbit antiserum (dilution 1:10 000) raised against purified allergen Pru p 3 (peach LTP) by immunizing rabbits. As a negative control, a strip with transferred peel extract was incubated with rabbit preimmune serum at the same dilution. After washing, the strips were incubated with peroxidase-conjugated goat antirabbit antibody (GAR-PO, Calbiochem) diluted 1:20 000. Finally, the strips and the proteins were washed, and IgE-binding proteins were detected using chemoluminescence (ECL, GE Healthcare) [3].

The figure shows the result of immunoblotting with polyclonal anti-LTP rabbit serum. Two bands are visible. One has a molecular weight of approximately 11 to 12 kDa and corresponds to the molecular weight reported for the LTPs; the other has a high molecular weight that could be attributed to high-molecular-weight aggregates.

Allergy to persimmon is uncommon. There have been reports of skin rash [4], urticaria, asthma [5], and even anaphylaxis [6,7] after ingestion. Previous studies on allergy to persimmon describe the involvement of various panallergens, such as the major allergen of birch pollen (Bet v 1) [8], profilin (Bet v 2), and carbohydrate determinants [5], suggesting primary sensitization to pollen or latex. However, the patient in the present case did not have sIgE to profilin, despite having rhinoconjunctivitis due to grass pollen. He did, however, have high sIgE values against peach LTP. The immunology workup revealed that the patient's serum recognized a band of approximately 12 kDa (coinciding with that of LTP) that only appeared in the peel. Different degrees of sequence identity have been found for LTP among family members of different species [9]. These range from 30% to 95%, although in the present case, given that immunoblotting inhibition of Pru p 3 was unable to inhibit binding of the patient's serum to the persimmon band, there may not have been sufficient structural and sequence identity between peach LTP and persimmon LTP. This possibility is consistent with the observation that the patient only developed symptoms with persimmon and tolerated other fruits.

Finally, the use of polyclonal anti-LTP rabbit serum seems to demonstrate the presence of LTP in the peel.

To our knowledge, this is the first case of selective allergy to persimmon in which the results of the in vitro study revealed sensitization to persimmon LTP as a possible cause of the reaction.

Funding

The authors declare that no funding was received for the present study.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

References

1. Towbin H, Staehelin Y, Gordon J. Electrophoretic transfer of proteins from polyacrylamide gels to nitro-cellulose sheets: procedure and some applications. Proc Natl Acad Sci USA. 1979;76:4350-4.

2. Sánchez-Madrid F, Morago G, Corbi AL, Carreira J. Monoclonal antibodies to three distinct epitopes on human IgE: their use for determination of allergen-specific IgE. J Immunol Methods. 1984;73:367-78.
3. Duffort OA, Polo F, Lombardero M, Díaz-Perales A, Sánchez-Monge R, García-Casado G, Salcedo G, Barber D. Immunoassay to quantify the major peach allergen Pru p 3 in foodstuffs. Differential allergen release and stability under physiological conditions. J Agric Food Chem. 2002;50:7738-41.
4. Kitano A, Miyazaki T, Yoshioka K, Kurono T, Kurono S, Matsumoto T. Facial rash and palmoplantar pruritus in an infant after first contact with kaki. J Investig Allergol Clin Immunol. 2009;19:237-8.
5. Anliker MD, Reindl J, Vieths S, Wüthrich B. Allergy caused by ingestion of persimmon (Diospyros kaki): detection of specific IgE and cross-reactivity to profilin and carbohydrate determinants. J Allergy Clin Immunol. 2001;107:718-23.
6. Martínez JC, Armentia A, Bartolomé B, Callejo A, Fuentes MJ, Fernández A. Anaphylaxis after ingestion of sharon fruit. Allergol Immunopathol. 2001;29:69-71.
7. Prandini M, Marchesi S. Anaphylaxis to persimmon. Allergy. 1999;54:897.
8. Bolhaar ST, van Ree R, Ma Y, Bruijnzeel-Koomen CA, Vieths S, Hoffmann-Sommergruber K, Knulst AC, Zuidmeer L. Severe allergy to sharon fruit caused by birch pollen. Int Arch Allergy Immunol. 2005;136:45-52.
9. Salcedo G, Sanchez-Monge R, Diaz-Perales A, Garcia-Casado G, Barber D. Plant non-specific lipid transfer proteins as food and pollen allergens. Clin Exp Allergy. 2004;34:1336-41

Manuscript received October 11, 2013; accepted for publication, May 28, 2014.

Beatriz Rodríguez Jiménez
Unidad de Alergología
Hospital Universitario de Getafe
Carretera de Toledo Km 12,500
28905 (Getafe) Madrid, Spain
E-mail: brodriguezjimenez@hotmail.com

Occupational Asthma to Dried Tobacco Leaves: A Very Delayed Diagnosis

Penven E[1,2], Poussel M[3,4], Thaon I[1,2], Paris C[1,2]
[1]Occupational Diseases Department, Bâtiment Philippe Canton, CHU Nancy, Vandoeuvre-lès-Nancy, France
[2]EA-7298 INGRES, Université de Lorraine, Vandoeuvre-lès-Nancy, France
[3]Department of Pulmonary Function Testing and Exercise Test, CHU Nancy, Vandoeuvre-lès-Nancy, France
[4]EA 3450 DevAH - Development, Adaptation and Disadvantage, Cardiorespiratory regulations and motor control, Université de Lorraine, Vandoeuvre-lès-Nancy, France

Key words: Occupational asthma. Tobacco. Specific inhalation challenge.

Palabras clave: Asma ocupacional. Tabaco. Prueba específica de provocación por inhalación.

A 59-year-old woman who had never smoked and had no known history of atopy was seen in our department in 2011 for a possible diagnosis of occupational asthma. The patient had not been employed since 2000 and had worked as a production agent at a cigarette manufacturing facility between 1986 and 2000. Her work primarily consisted of manually filling small bags with dried, milled tobacco leaves. She described the factory as very dusty, particularly during the early years, but no atmospheric measurements were available. The patient reported the appearance of rhinitis, cough, dyspnea, and wheezing, closely related to work periods, some months after starting to work at the factory. An initial check-up in 1991 led to a diagnosis of asthma, but a skin prick test (SPT) to tobacco leaves yielded a wheal of just 2 mm in diameter and was considered doubtful. The patient continued to work until 2000 without any change in her exposure to tobacco leaves; she described progressive worsening of her asthma, despite short-acting β_2-agonist treatment. The patient stopped working at the factory in 2000 and was no longer exposed to respiratory allergens or irritants. Her respiratory symptoms decreased, but did not disappear completely. In 2008, she experienced worsening of dyspnea and received inhaled corticosteroid therapy, with only slight improvement due to poor treatment adherence. In 2011, the patient was referred to our department for a possible diagnosis of occupational asthma. Clinically, she had bronchial hyperresponsiveness, but no wheezing under treatment. SPTs to airborne allergens and tobacco leaves (after humidification) were negative. The blood count was normal and total IgE was 1451 IU/mL. Specific IgE levels were 0.21 kU_A/L for tobacco leaves and 0.12 kU_A/l for eggplant. The results were negative for latex, tomato, and potato allergens. The baseline functional respiratory test demonstrated a slight reversible obstructive syndrome (forced expiratory volume in the first second [FEV_1], 2.11 L; 91% of predicted; FEV1/forced vital capacity [FVC],70%; forced expiratory flow at 50% relative to FVC, 1.97 L/s; 54% of predicted). A methacholine challenge was

positive with a 36% decrease in FEV_1 for a cumulative dose of 160 µg of 1% methacholine (approximately 0.5 mg/mL). An inhalation control test to lactose powder (stepwise handling of lactose powder for 1, 2, 3, 4 and 5 minutes) was strongly negative. In the specific inhalation challenge (SIC) to tobacco, the patient was asked to pour 2 cups of 20 g of tobacco leaf powder according to the same schedule as that used for lactose powder. A strong immediate positive reaction appeared after 10 minutes of cumulative exposure, with a 42% decrease in FEV_1 relative to baseline. No delayed symptoms were observed before discharge from our department 8 hours after the SIC, but the general practitioner reported some wheezing the following morning. To rule out nonspecific irritant reactions, we performed SICs with tobacco leaves in the same conditions in 2 volunteers, using a positive methacholine test as a control. These 2 SICs were strongly negative. A possible diagnosis of occupational asthma to tobacco leaves was established.

Occupational asthma to tobacco dust was first described by Gleich et al [1] in 1980. Since then, many authors have reported cases of occupational asthma as well as alterations in respiratory function in cigarette facilities. In 1988, for instance, Lander et al [2] reported a significant change in daytime peak flow expiratory in tobacco workers compared with controls. More recently, Mustajbegovic et al [3], following the systematic examination of 121 tobacco workers, reported 6 cases of occupational asthma to tobacco dust, interestingly all in women (total women, 97). To date no tobacco allergens have been identified. Although contamination of tobacco by fungi was initially hypothesized, more recent findings suggest that a profilin-like protein, or a villin-like protein [4] belonging to the cytoskeletal of plants, may be involved, as there have been several (but inconsistent) reports of cross-reactivity between several allergens from the Solanaceae family [5-6] as well as latex [7] in individuals with tobacco leaf asthma. Although our case is consistent with previously reported cases, the diagnosis of occupational asthma is questionable considering that the source of occupational exposure was eliminated a long time ago. The absence of atopy or previous asthma, intense occupational exposure to tobacco leaves for more than 10 years, and the patient's clinical history all support this potential diagnosis but may not be sufficient. The strongest diagnostic evidence is the positive SIC to tobacco leaf powder. However, this may also correspond to a simple immediate reaction to a nonspecific irritant. The absence of a reaction to the control test to lactose powder using the same procedure, the severity of the specific response (fall of 42% in FEV_1 relative to baseline), and the existence of a slight delayed reaction at 24 hours all support a diagnosis of occupational asthma rather than a simple immediate irritant reaction to tobacco leaf dust. Nevertheless, specific IgE to tobacco leaves was low, but this might be explained by the long period without exposure. We therefore believe that a diagnosis of occupational asthma to tobacco leaves is the most plausible diagnosis. Consequently, even though end of exposure is often proposed as an explanation for a negative SIC, our observation suggests that positive reactions may still occur many years later. Clinicians should also be aware that functional respiratory reactions could still be severe in such cases.

Funding

The authors declare that no funding was received for the present study.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

References

1. Gleich GJ, Welsh PW, Yunginger JW, Hyatt RE, Catlett JB. Allergy to tobacco: an occupational hazard. N Engl J Med 1980;302:617-9.
2. Lander F, Gravesen S. Respiratory disorders among tobacco workers. Br J Ind Med 1988;45:500-2.
3. Mustajbegovic J, Zuskin E, Schachter EN, Kern J, Luburic-Milas M, Pucarin J. Respiratory findings in tobacco workers. Chest 2003;123:1740-8.
4. Mittermann I, Voronin V, Heberle-Bors E, Valenta R. Identification of a villin-related tobacco protein as a novel cross-reactive plant allergen. FEBS Lett 2005;579:3807-13.
5. Armentia A, Bartolomé B, Puyo M, Paredes C, Calderón S, Asensio T, del Villar V. Tobacco as an allergen in bronchial disease. Ann Allergy Asthma Immunol 2007;98:329-36.
6. Ortega N, Quiralte J, Blanco C, Castillo R, Alvarez MJ, Carrillo T. Tobacco allergy: demonstration of cross-reactivity with other members of Solanaceae family and mugwort pollen. Ann Allergy Asthma Immunol 1999;82:194-7.
7. Armentia A, Dueñas-Laita A, Bartolomé B, Martín-Gil FJ, San Miguel A, Castrodeza JJ. Clinical significance of cross-reactivity between tobacco and latex. Allergol Immunopathol (Madr) 2010;38:187-96.

Manuscript received February 14, 2014; accepted for publication, June 2, 2014.

Christophe Paris
Centre de consultations de pathologies professionnelles
CHU Nancy
Bâtiment Philippe Canton
Rue du Morvan
Vandoeuvre-lès-Nancy, F-54511, France
E-mail: christophe.paris@inserm.fr

Not All Facial Swellings Are Angioedemas!

Fricker M[1], Dubach P[2,3], Helbling A[1], Diamantis E[4], Villiger PM[1], Novak U[5]

[1]*Department of Rheumatology, Clinical Immunology and Allergology, Inselspital, Bern University Hospital and University of Bern, Bern, Switzerland*

[2]*Department of ENT, Head and Neck Surgery, Inselspital, Bern University Hospital and University of Bern, Bern, Switzerland*

[3]*Innovation Center for Computer Assisted Surgery University of Leipzig, Germany*

[4]*Institute of Pathology, University of Bern, Switzerland*

[5]*Department of Medical Oncology, Inselspital, Bern University Hospital and University of Bern, Bern, Switzerland*

Key words: Angioedema. Facial swelling. Panniculitis. Subcutaneous panniculitis-like T-cell lymphoma. Chemotherapy.

Palabras clave: Angioedema. Inflamación facial. Paniculitis. Linfoma subcutáneo de células T tipo paniculitis. Quimioterapia.

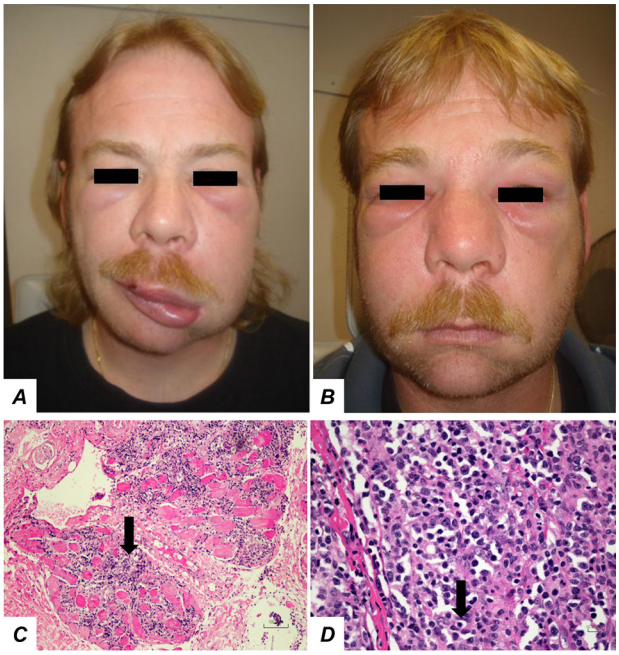

Figure. Swelling of the left part of the face at presentation (A) and when chemotherapy was initiated (B). C, Atypical lymphoid cells diffusely infiltrating the deep subcutaneous tissue (arrow) and the skeletal muscle (hematoxylin-eosin, x100). D, The neoplastic cells show round to oval nuclei with inconspicuous nucleoli and pale cytoplasm (hematoxylin-eosin, x400).

We report the case of a 38-year old man referred to our clinic with a fluctuating swelling of the face over the lower jaw. He was a white farmer and truck driver with an unremarkable past medical history. An initial nuclear magnetic resonance imaging (MRI) scan of the neck and head region showed a moderate dermal infiltration without necrosis or bone destruction lateral to the horizontal mandibular ramus. The fluctuating swelling was painless and was not associated with exanthemas or constitutional symptoms such as fever, malaise, weight loss, or arthralgia. Symptomatic treatment with antihistamines and corticosteroids was ineffective. No triggers such as drugs, foods, or specific contact substances were identified. Clinical examination showed a grossly disfiguring and indurated pasty swelling of the left lower lip and cheek (Figure A). Laboratory findings were normal (including differential blood counts, C-reactive protein, liver enzymes, C1-esterase inhibitors, C4, baseline tryptase, and flow-cytometric analysis of T- and B-cell subpopulations). Fine-needle aspiration and deep biopsies were inconclusive and in particular showed no signs of dysplasia or malignancy. Given the presence of a single epithelioid cellular granuloma, an early form of a necrotizing sialometaplasia was hypothesized as part of the differential diagnosis. Mycobacterial infections, including tuberculosis, were ruled out by direct staining and cultures. A 2-week course of empirical antibiotics with amoxicillin/clavulanic acid was ineffective. Based on the clinical presentation and the histological detection of a solitary granuloma, both cheilitis granulomatosa (Melkersson-Rosenthal syndrome) and local sarcoidosis were considered. Surprisingly, the swelling regressed spontaneously, but recurred within months, at which time it also affected the subcutis of the mid face extending down to the lower margin of the mandible, predominantly on the left side. A second MRI did not reveal abscess-forming processes or a focal nodular component. Because of a suspected soft tissue panniculitis (data not shown), combined treatment with clindamycin for 3 weeks and oral corticosteroids (initial dose of 100 mg for 5 days tapered down to 0 mg) over 1 month was prescribed, again without any clinical benefit. The patient could not be convinced to undergo reassessment with a repeat biopsy. An allergological workup with commercial prick test solutions with common aeroallergens (ALK-Abello) revealed sensitizations to rat epithelium, house dust mites, and *Lepidoglyphus destructor* (a storage mite species); these were all of questionable relevance given the absence of respiratory symptoms. Laboratory findings at the time displayed moderate absolute lymphopenia (993 cells/µL; range, 1200-2800 cells/µL) with a slight decrease in CD4 lymphocytes (225 cells/µL; range, 410-1590 cells/µL). Possible infection from the patient's pet—a rat—was discussed with the patient in reference to the report "Rat Bite: An Unusual Cause of Orbital Cellulitis" [1], and thus, an orofacial syndrome of unknown origin was the working hypothesis at the time.

Over the following months, the indurated, nontender, and nonulcerating swelling progressed to involve the whole left face accompanied by paresis of the marginal branch of the facial nerve and significant periorbital lymphedema (Figure B). The patient now agreed to another biopsy, which consisted of deep incisional biopsies of the cheek and the nasal vestibule. Histological analyses revealed the final diagnosis of a subcutaneous panniculitis-like T-cell lymphoma (SPTL) 1 year after the first signs of disease (Figure C, D). This entity, first described by Gonzalez et al [2] in 1991, is a rare T-cell lymphoma of the subcutaneous

tissue that clinically mimics panniculitis. It is associated with diverse autoimmune disorders in approximately 20% of patients and, compared with other cutaneous lymphomas, is often characterized by aggressive clinical behavior [2,3]. A retrospective analysis of a cohort of 83 cases proposed that SPTL may harbor 2 distinct entities with a different T-cell immunophenotype, clinical presentation, and prognosis [4]. Standard staging procedures including a whole-body computed tomography scan and bone marrow biopsies revealed no other lymphoma manifestations or signs of hemophagocytosis. The patient was given 6 uneventful cycles of chemotherapy with cyclophosphamide, doxorubicin, vincristine, and prednisolone (standard CHOP regimen) [4,5]. The treatment led to rapid, complete regression of the swelling, which, clinically, further uncovered the paresis of the facial nerve. This paresis has not yet fully resolved, despite several month of speech therapy.

The present case clearly demonstrates that a persistent and progressive swelling, atypical for angioedema or other known immunological or rheumatological disorders and unresponsive to treatment, needs further unbiased investigations including repeat biopsies. Diagnosis of SPTL is often delayed, for various reasons, for up to 10 years [4]. Forcing an earlier diagnosis may have improved the outcome of the facial palsy in our patient. Given its clinical presentation mimicking panniculitis in various locations and the association with autoimmune disorders, this rare lymphoma may be seen in early stages by rheumatologists or clinical immunologists [3], especially in the absence of ulcerations.

Acknowledgments

The authors wish to acknowledge Franziska Mitton Schmid for her valuable and continuous administrative support.

Funding

The authors declare that no funding was received for the present study.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

References

1. Ouazzani BT, Dali H, Daoudi R, Chakir M, Jiddane M, Mohcine Z: [Rat bite: an unusual cause of orbital cellulitis]. J Fr Ophtalmol. 2006;29:e14.
2. Gonzalez CL, Medeiros LJ, Braziel RM, Jaffe ES: T-cell lymphoma involving subcutaneous tissue. A clinicopathologic entity commonly associated with hemophagocytic syndrome. Am J Surg Pathol. 1991;15:17-27.
3. Velez NF, Ishizawar RC, Dellaripa PF, Saavedra AP, Laga AC, Murphy GF, Fisher DC, Kupper TS, Vleugels RA: Full facial edema: a novel presentation of subcutaneous panniculitis-like T-cell lymphoma. J Clin Oncol. 2012;30:e233-6.
4. Willemze R, Jansen PM, Cerroni L, Berti E, Santucci M, Assaf C, Canninga-van Dijk MR, Carlotti A, Geerts ML, Hahtola S, Hummel M, Jeskanen L, Kempf W, Massone C, Ortiz-Romero PL, Paulli M, Petrella T, Ranki A, Peralto JL, Robson A, Senff JN, Vermeer MH, Wechsler J, Whittaker S, Meijer CJ; EORTC Cutaneous Lymphoma Group. Subcutaneous panniculitis-like T-cell lymphoma: definition, classification, and prognostic factors: an EORTC Cutaneous Lymphoma Group Study of 83 cases. Blood. 2008;111:838-45.
5. Gallardo F, Pujol RM: Subcutaneous panniculitic-like T-cell lymphoma and other primary cutaneous lymphomas with prominent subcutaneous tissue involvement. Dermatol Clin. 2008;26:529-40,viii.

Manuscript received February 6, 2014; accepted for publication, June 5, 2014.

Urban Novak
INSELSPITAL, Universitätsspital Bern
Klinik und Poliklinik für Medizinische Onkologie
CH-3010 Bern, Switzerland
E-mail: urban.novak@insel.ch

First Case Report of Acute Generalized Exanthematous Pustulosis Due to Labetalol

Gómez Torrijos E[1], García Rodríguez C[1], Sánchez Caminero MP[2], Castro Jiménez A[1], García Rodríguez R[1], Feo-Brito F[1]
[1]*Allergy Section, Hospital General Universitario, Ciudad Real, Spain*
[2]*Dermatology Section, Hospital General Universitario, Ciudad Real, Spain*

Key words: Acute generalized exanthematous pustulosis. Delayed hypersensitivity. Labetalol. β-Blockers. Atenolol.

Palabras clave: Pustulosis exantemática aguda generalizada. Hipersensibilidad retardada. Labetalol. Fármacos betabloqueantes. Atenolol.

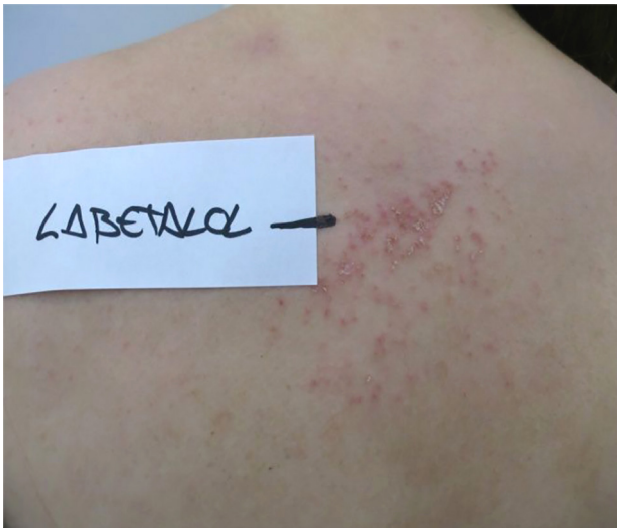

Figure. Positive patch test results with labetalol 5% (48 hours).

Most adverse drug reactions have a specific clinical pattern, and it is often impossible to identify the causative agent, especially when the patient is taking multiple drugs simultaneously. Management of an adverse reaction is based on a complete clinical history and a detailed study of the possible causative drug with skin tests, in vitro tests, and/or oral challenge tests.

A 31-year-old patient with a history of hypertension in the third trimester of pregnancy was treated with labetalol for 18 days and hydralazine α-methyldopa and metamizole during the 3 days immediately preceding admission. She denied any personal or family history of psoriasis or allergy to inhalants or drugs. About 8 hours before cesarean delivery of twins, erythematous micropapular lesions appeared on the face and neck. These became generalized in 4-5 days, affecting flexures, the intermammary cleft, chest, back, and palms and soles, with multiple micropustules accompanied by mild dermal itching and discomfort when swallowing. Physical examination revealed pharyngeal enanthem, no lymphadenopathy or organ enlargement, and low-grade fever (38.7°C). Despite withdrawing α-methyldopa, metamizole, and hydralazine on the second day of the eruption, new lesions continued to appear. On the fifth day after discontinuing labetalol, no new skin lesions had appeared.

The blood sample analysis disclosed the following values: leukocytes, 20 300/μL (90% neutrophils, 5% lymphocytes, and 5% monocytes); erythrocytes, 4 390 000/μL; hemoglobin, 14 g/dL; hematocrit, 42%; platelets, 145 000/μL; erythrocyte sedimentation rate, 30 mm/h; aspartate aminotransferase, 37 IU/L; alanine aminotransferase, 72 IU/L; γ-glutamyl transpeptidase, 125 IU/L; and alkaline phosphatase, 234 IU/L. Culture of the pustule content was negative. The results of viral serology were as follows: Epstein-Barr VCA P18 IgG (capsid), positive; Epstein-Barr gp-125 IgM (capsid), negative; Epstein-Barr EBNA IgG antinuclear antibody, positive; rubeola, immune. Negative values were found for parvovirus G19 (IgM, 0.33; IgG, 0.32), *Toxoplasma gondii* (IgG), and cytomegalovirus (IgM).

Patch tests performed 1 month after the onset of symptoms with labetalol, metamizole, hydralazine, and α-methyldopa (5% in water and petrolatum) were positive only for labetalol (water and petrolatum), and pustules were observed on the area tested (Figure). The results of challenge testing with metamizole, α-methyldopa, and hydralazine were all negative, with good tolerance.

In order to offer an alternative to labetalol, patch tests were performed with 10% atenolol in water and petrolatum, and the results were negative. The result of the subsequent tolerance test with atenolol, however, was positive, and 1 hour after taking 25 mg, the patient complained of generalized itching and micropapules on her back and palms, which persisted for up to 48 hours despite treatment with 60 mg of 6-methylprednisolone. The dose was reduced to 16 mg every 8 hours for 2 days.

A skin biopsy of the pustules showed slightly acanthotic skin with subcorneal pustules marking the surface, underlying spongiosis, and spongiosis elsewhere in the epidermis. Polymorphonuclear exocytosis was also observed. A mild lymphohistiocytic inflammation with some interstitial neutrophils (periodic acid-Schiff–negative) was seen in the superficial dermis.

Consequently, the patient was diagnosed with acute generalized exanthematous pustulosis (AGEP) due to labetalol with cross-reactivity to atenolol.

AGEP is an acute follicular rash that manifests with very small pustules on an erythematous edematous base that appear on the face, neck, and flexures 1-2 days after exposure to the offending drug, before rapidly becoming generalized. It is associated with fever and, sometimes, systemic symptoms, which resolve spontaneously in about 2 weeks. The condition is characterized by intradermal spongiform pustules, edema of the papillary dermis, and predominantly perivascular inflammatory infiltrate, which is occasionally associated with leukocytoclastic vasculitis [1].

Ninety percent of cases of AGEP are due to drugs such as β-lactam antibiotics and macrolide antimicrobials (hydroxychloroquine, cotrimoxazole terbinafine,

metronidazole, nystatin, and chloroquine), diltiazem, paracetamol, celecoxib, carbamazepine, povidone iodine, allopurinol, metformin, sildenafil, spiramycin, abacavir, and quinolones [2-4]. Other less common causes include viral infections by enterovirus, cytomegalovirus, and parvovirus B19 [5], exposure to mercury, and ingestion of food allergens [6]. In the case we present, the patient had begun treatment with labetalol 18 days earlier. She probably became sensitized during this period, as the latency period after the second exposure to another cross-reactive drug was only 1 hour [7]. Although the increased latency period is greater than that described in other cases of AGEP, it can sometimes reach 3-4 weeks, as in other types of drug eruption (eg, drug reaction with eosinophilia and systemic symptoms). The interest of the present case lies in the fact that, even though the patient was treated with several drugs, the allergy workup and the skin biopsy provided abundant detail. Therefore, biopsy confirmed the diagnosis, the patch tests identified labetalol as the causative drug (but failed to detect cross-reactivity with other β-blockers), and an oral tolerance test with atenolol was positive.

The EuroSCAR (Severe Cutaneous Adverse Drug Reactions) group has reported the diagnostic criteria for this condition [7], namely, acute pustular rash, fever >38°C, neutrophilia with or without eosinophilia, subcorneal or intraepidermal pustules in the skin biopsy, and spontaneous resolution in <15 days.

We performed a differential diagnosis with generalized pustular psoriasis, which showed that our patient had no history of psoriasis, duration was similar to that described in AGEP, and the course of generalized pustular psoriasis is longer. We also ruled out acute generalized pustulosis, which primarily affects children and adolescents, has a predominantly acral distribution, and is believed to be due to streptococcal infection [8].

The diagnostic value of patch testing in AGEP varied in different series, with positive results observed in between 32% and 50% of cases [9]. However, its utility is limited to cases with positive. Negative results do not rule out the involvement of a specific drug; therefore, intradermal tests with the drug involved and a late reading are recommended.

AGEP usually has a good prognosis, with a mortality rate of around 5%, and resolves spontaneously upon removal of the offending drug [10]. In cases of associated extensive systemic skin involvement, support measures may be needed. Treatment with systemic corticosteroids is not generally considered necessary. Reintroduction of the offending drug should be avoided, owing to the risk of recurrence, which is usually characterized by a more rapid onset (a few hours).

We report the first case of AGEP due to labetalol and cross-reactivity with atenolol.

Funding

The authors declare that no funding was received for the present study.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

References

1. Machet L, Martin L, Vaillant L. Acute generalized exanthematous pustulosis. Ann Dermatol Venereol. 2001;128:73-9.
2. Solla Babío E, Suárez Amor OM, Pérez Valcárcel J. Pustulosis exantematica generalizada aguda inducida por amoxicilina-clavulánico Galicia Clin. 2010;71 (4):185-6.
3. Jean-Claude Roujeau JC, Bioulac-Sage P, Bourseau C, Guillaume JC, Bernard Ph, Catherine Lok C, Plantin P, Claudy A, Delavierre C,Vaillant L, Wechsler J, Danan G, Bénichou C, Beylot C, Acute generalized exanthematous pustulosis. Analysis of 63 cases. Arch Dermatol. 1991;127:1333-8.
4. Saissi EH, Beau-Salinas F, Jonville-Béra AP, Lorette G, Autret-Leca E. Drugs associated with acute generalized exanthematic pustulosis. Ann Dermatol Venereol. 2003;130:612-8.
5. Ofugi S, Yamamoto O. Acute generalized exanthematous pustulosis associated with a human parvovirus B19 infection. J Dermatol. 2007;34:121-3.
6. Park YM, Park JG, Kang H, Houh D, Byun DG, Kim JW. Acute generalized exanthematous pustulosis induced by ingestion of lacquer chicken. Br J Dermatol. 2000;143:198-200.
7. Sidoroff A, Halevy S, BouwesBavinckJN, Vaillant L, Roujeau JC. Acute generalized exanthematous pustulosis (AGEP): a clinical reaction pattern. J Cutan Pathol. 2001;28:113-9.
8. Auer-Grumbach P, Pfaffenthaler E, Soyer H. Pustulosis acutageneralisata is a poststreptococcal disease and is distinct from acute generalized exanthematous pustulosis. Br J Dermatol. 1995;133:135-9.
9. Barbaud A. Drug patch testing in systemic cutaneous drug allergy. 2005;209:209-16.
10. Roujeau JC. Clinical heterogeneity of drug hypersensitivity. Toxicology. 2005;209:123-9.

Manuscript received April 29, 2014; accepted for publication June 6, 2014.

Elisa Gómez Torrijos
Sección de Alergología
Hospital General Universitario de Ciudad Real
C/ Obispo Rafael Torija, s/n
13005 Ciudad Real, Spain
E-mail: egomezt.cr@gmail.com

Prevalence of Sensitization to Pollen From Trees Planted in Barcelona City

Puiggròs A[1], Muñoz-Cano R[2], Roger Reig A[3], Raga E[4], Belmonte J[5], Valero A[2], on behalf of Comitè d'Al.lergia Respiratòria de la Societat Catalana d'Al·lergologia i Immunologia Clínica (SCAIC) Catalunya, Spain (Asensio de la Cruz O, Eseverri Asín JL, Güell Figueras E, San Miguel Moncín MM, Torredemer Palau A, Bartra J, Tella R, Sala-Cunill A, Dalmau G)

[1]*Allergy Unit, Hospital Quiron, Barcelona, Catalonia, Spain*

[2]*Allergy Unit, Pneumology and Allergy Department, Hospital Clinic-IDIBAPS, Centro de Investigaciones Biomédicas en Red de Enfermedades Respiratorias (CIBERES), Barcelona, Catalonia, Spain*

[3]*Centre Roger Barri d'Asmologia i Al.lèrgia, Barcelona, Catalonia, Spain*

[4]*Allergy Unit, Hospital Plató, Barcelona, Catalonia, Spain*

[5]*Botany Unit and Institute of Environmental Science and Technology (ICTA), Universitat Autònoma de Barcelona, Bellaterra, Spain*

Key words: Rhinitis. Asthma. Airborne tree pollen. Sensitization prevalence. Barcelona.

Palabras clave: Rinitis. Asma. Polen atmosférico. Prevalencia de sensibilización. Barcelona.

Allergic rhinitis is an increasingly important disease because of its prevalence (30% worldwide), its effect on patients' quality of life, and its coexistence with other diseases such as asthma and conjunctivitis [1-3]. According to the epidemiological study Alergológica 2005 [4], sensitization to *Platanus hispanica* was the most important cause of allergic rhinitis due to pollen. Some studies have demonstrated a correlation between symptoms and pollen levels [5].

The aim of the study was to determine the allergenic profile of the various species of tree planted in the streets of Barcelona, Catalonia, Spain.

A prospective, multicenter, epidemiological study was performed between April 2008 and October 2010. The study population comprised adult patients with respiratory disease (asthma and/or rhinitis) attending an allergy unit for the first time. The participating centers were all located in Barcelona (Alergocentre, Centre Roger Barri d'Asmologia i Al.lèrgia, Fundació Sanitària Sant Pere Claver, Hospital de Sant Pau i la Santa Creu, Hospital Clínic Provincial, Hospital Plató, Hospital Quirón, and Hospital Universitari Vall d'Hebron).

All patients signed an informed consent document. The study was approved by the Ethics Committee at Hospital Universitari Germans Trias i Pujol, Badalona, Catalonia, Spain.

Data were collected online using the website www.gtcar1.org

All patients underwent skin prick testing with 2 panels of aeroallergens. The first panel tested included the so-called GA(2)LEN Pan-European core skin prick test panel [6], excluding *P hispanica* (already included in the Barcelona tree panel).

The second panel included the most frequent tree pollens in Barcelona.

Information regarding the trees planted in Barcelona was obtained from the census of plants published in 2005 and updated in 2008 by the Parks and Gardens Department (Departament de Parcs i Jardins) of Barcelona City Council. Only species with more than 2500 trees planted were included. In decreasing order of abundance, the species were *P hispanica, Celtis australis, Sophora japonica, Ulmus pumila, Robinia pseudoacacia, Tipuana tipu, Brachychiton populneus, Populus* species, *Melia azedarach, Ligustrum lucidum*, and *Phoenix dactylifera*.

In a second phase, the airborne pollen concentration was evaluated for each of the listed species. Since *S japonica, B populneus, M azedarach*, and *T tipu* were excluded from the study, we can assume that their allergenicity was limited because pollen was not detected in the atmosphere.

Data were analyzed using the statistical package SPSS 16.0 (SPSS Inc). The sample size was calculated to achieve a precision of 5% with a 95%CI. A *P* value of .05 or less was considered statistically significant. A multivariate logistic regression analysis was performed.

We studied 427 patients (53% men) with a mean (SD) age of 37 (11.3) years. Most reported a family history (59%) and personal history (72%) of atopy. Rhinitis affected 67%, asthma 4%, and both asthma and rhinitis 29%.

The most frequently detected pollen sensitizations were *P hispanica* (37.0%) and *L lucidum* (21.9%). The least

Table. Classification of Tree Species in the City of Barcelona According to Relative Sensitization

Species	Sensitized Patients, %	Trees, %	Relative Sensitization[a]	Potential Allergenicity
Platanus hispanica	37.0	34.1	1.08	Intermediate
Celtis australis	5.9	11.6	0.51	Low
Ulmus pumila	5.0	4.4	1.14	Intermediate
Robinia pseudoacacia	6.1	3.9	1.56	Intermediate
Populus nigra	3.1	3.1	1.00	Intermediate
Ligustrum lucidum	21.9	2.2	9.95	High
Phoenix dactylifera	6.6	1.7	3.88	High

[a]Sensitized patients/number of trees

common were *Populus nigra* (3.1%), *U pumila* (5.0%), *C australis* (5.9%), and *P dactylifera* (6.6%).

In order to measure the allergenicity of the pollens tested, we calculated the relative sensitization, that is, the ratio between the number of sensitized patients and the number of trees. Therefore, a high relative sensitization ratio indicated highly allergenic pollen. Although the highest prevalence of sensitization was recorded for *P hispanica*, the highest relative sensitization ratios were recorded for *L lucidum, P dactylifera*, and *R pseudoacacia*. In other words, lower numbers of trees are able to induce more cases of allergic sensitization. We classified the trees as having pollen with high, moderate, and low allergenicity depending on their relative sensitization rate (Table).

Taking into consideration the airborne pollen records provided by the Xarxa Aerobiològica de Catalunya, during the study period (2007-2010), *P hispanica* pollen accounted for 28.6% of the global pollen count. *Oleaceae* pollen accounted for 5% of the total pollen; olive tree pollen in particular represented 4% of the annual total *Oleaceae* pollen count. Among the remaining pollens, *P nigra* accounted for 1% of the total pollen detected, *P dactylifera* 0.4%, *U pumila* pollen 0.3%, and *C australis* 0.1%. *R pseudoacacia* pollen was not detected in Barcelona, probably because the pollen is enclosed by the flower and is spread by insects. In addition, the pollen of this species is difficult to differentiate from *Quercus* pollen.

We found a positive and statistically significant association between atmospheric pollen levels and sensitization rate (r=0.68, *P*=.044).

The prevalence of sensitization to tree pollen among patients with respiratory allergy in the city of Barcelona ranges from 3% to 37%. The highest prevalence of sensitization (37%) was to *P hispanica*.

Besides the positive correlation between pollen levels and the percentage of sensitization, the number of trees of each species planted should also be considered when attempting to define the allergenicity of each of them (relative sensitization). Thus, *P hispanica* was no longer the first cause of sensitization and was replaced by *L lucidum, P dactylifera*, and *R pseudoacacia*.

Our study was limited by the fact that we did not evaluate the clinical significance of sensitization or the potential cross-reactivity between pollens. Consequently, further studies would be required to assess the clinical impact of this sensitization.

In summary, the allergenicity of airborne tree pollen in the city of Barcelona depends not only on the number of trees planted and the pollen count, but also on the potential allergenicity of each species.

As a result of this study, the municipal authorities now take allergenicity into account when selecting the species to be planted.

Acknowledgments

We are grateful to researchers from participating centers (Amat P, Dordal T, González V, Muñoz E, Luengo O, and Rueda M) for their help with patient selection. We thank Laboratorios LETI for providing the skin prick test panels. We are grateful to Dr Jordina Belmonte for help with the airbone pollen records and to Andrés Hidalgo for creating the online database.

Funding

This work was supported by the Catalan Society of Allergy and Clinical Immunology (SCAIC) and Laboratorios LETI.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

References

1. Leynaert B, Neukirch C, Demoly P, Bousquet J. Epidemiologic evidence for asthma and rhinitis comorbidity. J Allergy Clin Immunol. 2000;106:S201-5.
2. Annesi-Maesano I. Epidemiological evidence of the occurrence of rhinitis and sinusitis in asthmatics. Allergy. 1999;54 Suppl 57:7-13.
3. Bauchau V, Durham SR. Prevalence and rate of diagnosis of allergic rhinitis in Europe. Eur Respir J. 2004;24:758-64.
4. Navarro AM. Rinitis. In: Sociedad Española de Alergología e Inmunología Clínica, Schering-Plough, eds. Factores epidemiológicos, clínicos y socioeconómicos de las enfermedades alérgicas en España en 2005. Madrid: SEAIC. Schering-Plough; 2006. p. 107-32.
5. Frenz DA. Interpreting atmospheric pollen counts for use in clinical allergy: allergic symptomatology. Ann Allergy Asthma Immunol. 2001;86:150-7.
6. Heinzerling L, Frew AJ, Bindslev-Jensen C, Bonini S, Bousquet J, Bresciani M, Carlsen K-H, van Cauwenberge P, Darsow U, Fokkens WJ, Haahtela T, van Hoecke H, Jessberger B, Kowalski ML, Kopp T, Lahoz CN, Lodrup Carlsen KC, Papadopoulos NG, Ring J, Schmid-Grendelmeier P, Vignola AM, Wöhrl S and Zuberbier T. Standard skin prick testing and sensitization to inhalant allergens across Europe- a survey from the GALEN network. Allergy. 2005;60;1287-300.

Manuscript received May 18, 2014; accepted for publication June 16, 2014.

Anna Puiggròs
Plaça Alfons Comín num. 5
Barcelona
Spain
E-mail: laputxi@gmail.com

Chlorhexidine: A Retrospective Observational Study of a Potentially Life-threatening Molecule

Bubenhofer M[1], Fricker M[1,3], Weber-Mani U[4], Helbling A[1,2]
[1]*Allergy Unit, Clinic of Internal Medicine, Zieglerspital, Spital Netz Bern, Bern, Switzerland*
[2]*Division of Allergology, Department of Rheumatology, Immunology and Allergology, University Hospital Bern, Bern, Switzerland*
[3]*Private Practice, Mörigen, Switzerland*
[4]*Private Practice, Thun, Switzerland*

Key words: Chlorhexidine. Chlorhexidine allergy. Drug hypersensitivity. Anaphylaxis. Disinfectants.

Palabras clave: Clorhexidina. Alergia a clorhexidina. Hipersensibilidad a medicamentos. Anafilaxia. Desinfectantes.

The disinfectant chlorhexidine (CHX) is a constituent of many medical products (including catheter coatings) and cosmetics [1]. Allergic symptoms due to CHX have been reported in case series [2]. Reported reactions range from localized delayed reactions to acute systemic reactions (with generalized urticaria, acute bronchospasm, and hypotension) and anaphylaxis [3]. The prevalence of allergy to CHX is not known. The aim of this study was to assess the circumstances surrounding and severity of allergic reactions to CHX in patients referred to the 2 allergy units of the Canton Bern in Switzerland.

A database in the clinical information system Phoenix (CompuGroup Medical) was searched to find all diagnoses of CHX allergy in the Division of Allergology, University Hospital, Inselspital, Switzerland and the Allergy Unit, Zieglerspital, Switzerland from January 1, 2005 to December 31, 2012. More than 4000 patients per year were referred to the units, which have a catchment area of approximately 1 million inhabitants. Two patients from the same catchment area and time period (patients 9 and 10) who attended the private office of UW-M were added. CHX allergy was diagnosed based on the clinical history and a series of tests performed over the 6 months following the index event.

Skin prick tests and scratch tests were performed with 0.5% CHX digluconate in 0.9% saline solution, the product causing the index reaction (Hibitane, Globopharm AG; Merfen, Novartis Pharma Schweiz AG; Instillagel, Melisana AG), or individual constituents thereof. The tests were performed on the volar forearm [4,5]. Patch tests were placed on the upper back using IQ Ultra PT Test Chambers (Chemotechnique Diagnostics) fixed with Hypafix (BSN medical). The results were evaluated and scored according to the criteria of the International Contact Dermatitis Research Group [6].

CHX-specific IgE (sIgE), total IgE, and tryptase were determined using ImmunoCAP FEIA (Thermo Fisher Scientific). sIgE levels >0.35 kU_A/L (radioallergosorbent test class ≥1) were considered positive, and tryptase levels ≥11.4 µg/L were considered elevated [7].

Within this 8-year period, 14 of more than 32 000 patients in the database plus the 2 from the private office had allergy to CHX. Patient-specific information is shown in the Table. Allergy to CHX was detected during urological procedures in 4 cases, at the dentist's or dental hygiene office in 6 cases, perioperatively in 3 cases, and at home in 3 cases.

Of the 16 patients, 13 (81%) had an immediate reaction, with symptoms occurring in most cases within 30 minutes of contact with CHX. In 9 of the 13 patients (69%), the reaction was anaphylactic, and hypotension or shock was documented in 7 (54%). Seven of the 13 had previously undergone a medical intervention that was similar to that causing the allergic reaction (data not shown). The results of skin tests to CHX were positive in 10 of 13 patients (77%). Three patients had negative skin test results (patients 2, 6, and 9). Of the 11 patients tested for sIgE, all had significantly elevated levels with a mean of 4.45 kU_A/L (range, 0.78-15.8 kU_A/L). Tryptase levels on the index day exceeded the normal range in 3 of 4 tested patients, with a mean peak of 44.3 µg/L (range, 16.1-92.9 µg/L).

Three of the 16 patients had a delayed reaction with localized swelling and eczematous skin alterations 24 to 72 hours after contact with CHX. Two patients had positive skin test results, and in 1 patient a negative scratch test result became positive within 24 hours and persisted for days.

In 2013, we obtained feedback from 8 of the 16 patients. Five (63%) reported allergic symptoms after subsequent contact with CHX. One patient (patient 6), who had previously experienced a life-threatening reaction after insertion of a central venous catheter, experienced a second life-threatening reaction after the same procedure.

While 16 cases of CHX allergy within 8 years in a catchment area of 1 million inhabitants may seem low, it is important to remember that the reaction was immediate and potentially dangerous in 13 cases. Even more striking, about one-third of the patients experienced a subsequent allergic reaction to CHX despite having undergone an allergy workup. These findings suggest that allergy to CHX is not well recognized by medical staff or by patients. The fact that CHX is very often an adjuvant rather than the main constituent of a medical product may explain in part this lack of awareness. Despite the ubiquitous use of CHX, most reactions occurred in medical environments (one-third at the dentist's office). Insertion of urinary and central venous catheters and oral rinsing with CHX solutions were the main causes of severe CHX allergy. Alert procedures should be in place in the respective settings, and patients with suspected CHX allergy should be tested and advised. We found determination of sIgE to CHX to be the most sensitive approach. Intradermal tests may be an alternative, since Garvey et al [2] found them to be appropriate in 100% of cases [2]. In order to enhance general awareness, Swissmedic (Swiss Agency for Therapeutic Products) recently released a pharmacovigilance report on 18 allergic reactions due to CHX (anaphylaxis was recorded in 9 patients, 1 of whom died [www.swissmedic.ch/marktueberwachung]).

Elevated tryptase levels on the index day indicate mast cell activation [7]. In 1 of the 4 patients tested, the peak tryptase value did not exceed the normal range, thus suggesting that

Table. Patient Characteristics, Circumstances, Symptoms, and Diagnostic Tests

Patient No. (Age/Sex)	Circumstances/Activity	Symptoms	Time[a]	Skin Tests[b]	sIgE, kU_A/L (RAST CLASS)	Total IgE, kU/L	Baseline Tryptase, µg/L	Peak Tryptase, µg/L[c]	Feedback
	Immediate type								
1 (57/male)	Urinary catheterization with Endosmed lubricant application[d]	Generalized exanthema	2 min	SPT +, PT +	7.11 (3)	634	3.5		Same reaction after CHX contact
2 (54/male)	Mouthwash with CHX-containing solution	Generalized urticaria	90 min	SPT –	1.03 (2)		7.8		Same reaction after CHX contact
3 (41/female)	Hernioplasty surgery	Intraoperative anaphylaxis/hypotonia	20 min	SPT +++	1.00 (2)	38	3.6	5.3	
4 (59/male)	Dental pouch rinsing	Paroxysmal intermittent atrial fibrillation, unconsciousness (15 min)	2 min	SPT M +++	13.0 (3)	762	<1.0	24	No more contact with CHX
5 (78/male)	Urinary Instillagel application before TURP[e]	Massive itching/generalized urticaria	30-60 min	SPT NC, PT +	2.49 (2)	250	3.6		Same reaction after CHX contact
6 (49/female)	Central venous catheterization	Hypotonic shock, cyanosis (Spo_2 35%)	1 min	SPT –	2.10 (2)	252	3.5	92.9	Same reaction after central venous catheterization
7 (36/male)	Central venous catheterization	Hypotonic shock (Spo_2 < 50%)	<1 min	SPT +	1.10 (2)	168		16.1	
8 (64/male)	Urinary catheterization with Instillagel application	Generalized urticaria/dyspnea/ hypotension	5 min	SPT M ++, SPT Ins ++, SrT Ins ++	1.65 (2)	49	3.27		
9 (27/male)	Topical application of Merfen[f] on finger wound at home	Generalized urticaria, tachycardia, angioedema, dysphagia, decreased GCS	2-3 min	SrT M ++, SrT CHX –, SrT B –		44.9	2.88		Same reaction after CHX contact
10 (52/male)	Mouthwash during dental hygiene	Nausea, urticaria of upper part of body, conjunctivitis, generalized itching	<1 min	SrT ++ CHX 2%					No more contact with CHX
11 (76/male)	Urethral catheterization before urologic surgery	Generalized urticaria, face swelling, hypotension	15 min	SPT ++, SrT ++	2.86 (2)	290	6.4		
12 (20/female)	Cleaning of gingival wound with CHX-containing solution after gingival excision	Generalized urticaria, angioedema, dyspnea, angina pectoris	<1 min	SPT +++	0.78 (1)		3.96		
13 (73/male)	Mouth rinsing with Plak-Out[g]	Collapse, mechanical reanimation, generalized urticaria	>1 min	SPT +++	15.8 (3)		5.92		
	Delayed type								
14 (59/male)	Pouch rinsing after tooth extraction	Bilateral lid edema, dysphagia, dyspnea	<48 h	SrT M + (48 hours)		16.1	9.36		No more contact with CHX
15 (11/male)	Topical Merfen on pustule on forehead at home	Local angioedema, itching	<24 h	PT M ++, PT CHX –, SrT CHX +			5.99		
16 (29/male)	Topical Merfen on wound on forearm at home	Localized eczema, itching, papules	<24 h	PT M +++		17.4			

Abbreviations: B, benzoxonium chloride; CHX, chlorhexidine; GCS, Glasgow Coma Scale; Ins, Instillagel; M, Merfen; NC, not conclusive; PT, patch test; Spo_2, peripheral capillary oxygen saturation; SPT, skin prick test; SrT, scratch test; TURP, transurethral prostate resection.
[a]Interval from contact to symptoms.
[b]+ 3-5 mm, ++ 5-7 mm, +++ >7 mm.
[c]Tryptase level measured on the day of reaction.
[d]Endosmed-gel, 0.5 mg/g CHX.
[e]Instillagel, 0.25% CHX/2% lidocaine.
[f]Merfen, 0.5% CHX/0.1% benzoxonium chloride
[g]Plak-Out, 2 mg/g CHX/E122.

other key mediators or pathway mechanisms are involved in anaphylaxis [8].

Delayed reactivity to CHX may be underrepresented in our specialty since this kind of dermal reaction is often not reported to an allergy unit. Nevertheless, our data are in line with those from other reports indicating that CHX has little relevance in contact allergy [9]. This is surprising, as the maximum concentration of CHX in cosmetics allowed in the European Union is 0.3%, which is the same as the concentration in the disinfectant Merfen (Novartis Pharma Schweiz AG), the cause of allergy in 3 of the patients we studied. For both sensitization and allergic reaction to CHX, factors other than the concentration may be relevant, for example, route of application and disrupted skin barrier [1,2]. Very few cases of immediate-type reactions to CHX through presumably intact skin have been reported [10].

In conclusion, CHX allergy is rare but can be life-threatening and often occurs repeatedly. Most severe allergic reactions occurred after venous or urinary catheterization and interventions at the dentist's office. To avoid recurrence, medical staff should undergo appropriate training, and an allergology workup should be performed when CHX allergy is suspected.

Funding

The authors declare that no funding was received for the present study.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

Previous Presentation

An abstract of this article was presented as a poster at the 2013 SGAI Congress in Bern, Switzerland.

References

1. Calogiuri GF, Di Leo E, Trautmann A, Nettis E, Ferrannini A, Vacca A. Chlorhexidine hypersensitivity: A critical and updated review. J Allergy Ther. 2013;4:141.
2. Garvey LH, Kroigaard M, Poulsen LK, Skov PS, Mosbech H, Venemalm L, Degerbeck F. IgE-mediated allergy to chlorhexidine. J Allergy Clin Immunol. 2007;120:409-15.
3. Silvestri DL, McEnery-Stonelake M. Chlorhexidine: uses and adverse reactions. Dermatitis. 2013;24:112-8.
4. Brockow K, Garvey LH, Aberer W, Atanaskovic-Markovic M, Barbaud A, Bilo MB. Skin test concentrations for systemically administered drugs – an ENDA/EAACI Drug Allergy Interest Group position paper. Allergy. 2013;68:702-12.
5. Bousquet J, Heinzerling L, Bachert C, Papadopoulos NG, Bousquet PJ, Burney PG, Canonica, GW, Carlsen KH, Cox L, Haahtela T, Lodrup Carlsen KC, Price D, Samolinski B, Simons FER, Wickman M, Annesi-Maesano I, Baena-Cagnani CE, Bergmann KC, Bindslev-Jensen C, Casale TB, Chiriac A, Cruz AA, Dubakiene R, Durham SR, Fokkens WJ, Gerth-van-Wijk R, Kalayci O, Kowalski ML, Mari A, Mullol J, Nazamova-Baranova L, O'Hehir RE, Ohta K, Panzner P, Passalacqua G, Ring J, Rogala B, Romano A, Ryan D, Schmid-Grendelmeier P, Todo-Bom A, Valenta R, Woehrl S, Yusuf OM, Zuberbier T, Demoly P. Practical guide to skin prick tests in allergy to aeroallergens. Allergy. 2012;67:18-24.
6. Lachapelle J-M, Maibach HI. Patch testing methodology. In: Lachapelle J-M, Maibach HI, editors. Patch testing and prick testing: a practical guide - official publication of the ICDRG. 3rd ed. Berlin: Springer; 2012. p. 35-77.
7. Borer-Reinhold M, Haeberli G, Bitzenhofer M, Jandus P, Hausmann O, Fricker M, Helbling A, Müller U. An increase in serum tryptase even below 11.4 ng/mL may indicate a mast cell-mediated hypersensitivity reaction: a prospective study in Hymenoptera venom allergic patients. Clin Exp Allergy. 2011;41:1777-83.
8. Vadas P, Perelman B, Liss G. Platelet-activating factor, histamine, and tryptase levels in human anaphylaxis. J Allergy Clin Immunol. 2013;131:144-9.
9. Liippo J, Kousa P, Lammintausta K. The relevance of chlorhexidine contact allergy. Contact Dermatitis. 2011;64:229-34.
10. Heinemann C, Sinaiko R, Maibach H. Immunological contact urticaria and anaphylaxis to chlorhexidine: overview. Exog Dermatol. 2002;1:186-94

Manuscript received March 11, 2014; accepted for publication June 18, 2014.

Arthur Helbling
Division of Allergology, University Department of Rheumatology, Clinical Immunology & Allergology
University Hospital, Inselspital & Allergy Unit Zieglerspital
Department of Internal Medicine, Spital Netz Bern, Switzerland
E-mail: arthur.helbling@insel.ch
arthur.helbling@spitalnetzbern.ch

Fixed Drug Eruption Due to Atorvastatin

Huertas AJ[1], Ramírez-Hernández M[1], Mérida-Fernández C[1], Chica-Marchal A[2], Pajarón-Fernández MJ[1], Carreño-Rojo A[1]
[1]*Department of Allergy, University Hospital Complex of Cartagena, Murcia, Spain*
[2]*Department of Pharmacy, University Hospital Complex of Cartagena, Murcia, Spain*

Key words: Atorvastatin. Fixed drug eruption. Statins.

Palabras clave: Atorvastatina. Exantema fijo medicamentoso. Estatitnas.

Atorvastatin is a synthetic 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase inhibitor that is used to treat hypercholesterolemia. HMG-CoA reductase inhibitors are considered safe, and adverse reactions to statins are infrequent. We report a case of atorvastatin-induced fixed drug eruption.

An 84-year-old man was referred to our department from the dermatology department with a 7-month history of multiple erythematous macules extending from the trunk to the arms.

The patient had a history of hypercholesterolemia, diabetes mellitus, and arterial hypertension. He was under treatment with atorvastatin, metformin, trimetazidine, ramipril, clopidogrel, and omeprazole.

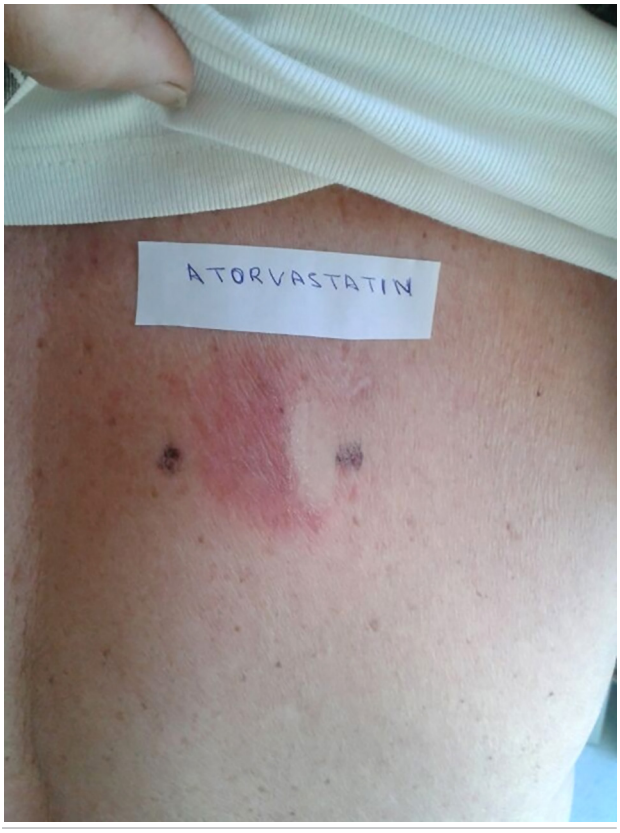

Figure. Positive patch test result with atorvastatin on a residual lesion.

Previous histological examination of the lesions in the dermatology department revealed chronic superficial dermatitis with infiltration of leukocytes, polymorphonuclear cells, and eosinophils in perivascular areas. This finding was compatible with fixed drug eruption.

Metformin was discontinued, and trimetazidine was stopped 6 weeks later. No changes were observed in the morphology of the skin lesions. Two months following withdrawal of atorvastatin, the erythema had partially resolved. The following month it had disappeared completely.

Closed patch tests were performed with atorvastatin and simvastatin 1/1000 in ethanol, as described elsewhere with simvastatin [1]. The result for atorvastatin was positive (++) at 96 hours on residual lesions (Figure). The result for simvastatin was positive at 48 hours (+) and at 96 hours (++) on residual lesions. The results were negative on healthy skin at 48 and 96 hours with both statins. We tested 6 controls, and the results for all of them were negative. These findings and the absence of lesions after withdrawal of atorvastatin confirmed the diagnosis of fixed drug eruption.

Fixed drug eruption is a distinctive drug eruption characterized by recurrent well-defined lesions at the same location each time the culprit drug is taken. Fixed drug eruption has been associated with many agents, including anticonvulsants, aspirin, nonsteroidal anti-inflammatory drugs, and antibiotics [2]. However, to our knowledge, fixed drug eruption due to atorvastatin or other statins has not been previously reported.

Few cases of hypersensitivity to atorvastatin have been reported to date. The reactions consisted of chronic urticaria [3], anaphylaxis [4], angioedema and eosinophilia [5], angioedema, dyspnea, nasal hydrorrhea [6], and drug reaction with eosinophilia and systemic symptoms syndrome [7]. In addition, atorvastatin has been implicated in other cutaneous adverse reactions such as toxic epidermal necrolysis [8], linear IgA bullous dermatosis [9], and psoriasis [10].

The patch test results with simvastatin in this report suggest cross-reactivity between different statins. Khan et al [4] reported the case of a patient who experienced anaphylaxis with atorvastatin and with simvastatin, thus supporting possible cross-reactivity between statins.

Funding

The authors declare that no funding was received for the present study.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

References

1. Peramiquel L, Serra E, Dalmau J, Vila AT, Mascaró JM, Alomar A. Occupational contact dermatitis from simvastatim. Contact Dermatitis. 2005;52:286-7.
2. Andrade P, Brinca A, Gonçalo M. Patch testing in fixed drug eruptions- a 20-years review. Contact Dermatitis. 2011;65:195-201.

3. Anliker MD, Wüthrich B. Cronic urticaria to artovastatin. Allergy. 2002;57:366-72.
4. Khan FS, Stewart DK, Brunzell JD, Natrajan KM, Castell MC, Henderson WR, Ayars AG. Successful desensitization to rosuvastatin in a patient a history of anaphylaxis to multiple statins. J Allergy Clin Immunol. 2013;131:234-6.
5. Hampson JP, Smith D, Cowell R, Baker A. Hypotension and eosinophilia with atorvastatin. Pharm World Sci. 2005;27:279-80.
6. Piñero Saavedra MC, Prados Castaño M, Sánchez B, Lucena Soto JM, Ávila Castellanos R, Ortega-Camarero M. Usefulness of lymphocyte activation test in atorvastatin hypersensitivity. J Investig Allergol Clin Immunol. 2011;21:571-7.
7. Gressier L, Pruvost-Balland C, Dubertret L, Viguier M. Atorvastatin-induced drug reaction with eosinophilia and systemic symptoms (DRESS). Ann Dermatol Venereol. 2009;136:50-3.
8. Pfeiffer CM, Kazenoff S, Rothberg HD. Toxic epidermal necrolysis from atorvastatin. JAMA. 1988;279:1613-4.
9. König C, Eickert A, Scharfetter-Kochanek K, Krieg T, Hunzelmann N. Linear IgA bullous dermatosis induced by atorvastatin. Am Acad Dermatol. 2001;44:689-92.
10. Cozzani E, Scaparro M, Parodi A. A case of psoriasis worsened by atorvastatin. J Dermatol Case Rep. 2009;30:60-1.

Manuscript received July 14, 2014; accepted for publication December 12, 2014.

M Ramírez-Hernández
Department of Allergy
University Hospital Complex of Cartagena
Paseo Alfonso XIII, 61
30203 Cartagena (Murcia)
Spain
E-mail: alergologa@hotmail.com

Aspirin Desensitization in a Patient With NSAID-Induced Delayed Angioedema

Rijo Y,[1] Canabal J,[1] Fiandor A,[1] Bobolea I,[2] Quirce S,[1] Cabañas R[1]
[1]*Hospital La Paz Institute for Health Research (IdiPaz), Department of Allergy, Madrid, Spain*
[2]*Hospital 12 de Octubre Institute for Health Research (i+12), Allergy Department, Madrid, Spain*

Key words: Aspirin. NSAID. Desensitization. Urticaria-angioedema. Coronary artery disease.

Palabras clave: Aspirina. AINE. Desensibilización. Urticaria-Angioedema. Enfermedad coronaria.

Aspirin (ASA) and other nonsteroidal anti-inflammatory drugs (NSAIDs) are the agents most commonly implicated in hypersensitivity reactions and are responsible for about 25% of all reactions. The most common clinical presentations are aspirin-induced asthma, rhinosinusitis, and aspirin-induced urticaria-angioedema. Symptoms usually appear immediately or a few hours after exposure to 1 or more NSAIDs and are thought to be IgE-mediated or associated with COX-1 inhibition, although some reactions are delayed (eg, severe bullous skin reactions), in which case the mechanism is thought to be T cell–mediated [1,2].

NSAIDs are widely used to treat many conditions and are often the main therapeutic option, even in hypersensitive patients. Induction of tolerance or desensitization is therefore recommended in patients who are likely to experience a hypersensitivity reaction [1,2].

Desensitization involves slow administration of increasing doses of ASA in order to reduce or eliminate potential pharmacological or immunological reactions [3]. It has proven successful in patients with ASA-induced urticaria-angioedema and ASA-exacerbated respiratory disease [4] but not in patients with chronic idiopathic urticaria [5,6]. There are no reports of desensitization in patients who experienced delayed skin reactions.

ASA is the first choice for prophylaxis in patients with coronary artery disease, since it has been shown to reduce mortality [7]. Ticlopidine and clopidogrel have been used as alternatives in patients with hypersensitivity to NSAIDs, although the cost-benefit ratio of these drugs has not been validated [8]. Dual antiplatelet therapy is indicated in acute coronary syndrome and in patients undergoing percutaneous coronary angioplasty and stent implantation. Desensitization enables long-term administration of ASA in these groups [4].

We report the case of a 56-year-old man with a history of chronic coronary artery disease. Catheterization was first performed in 2004, and subsequent management was noninvasive. Catheterization was performed again in 2011 with implantation of 3 stents. At the evaluation in the allergy department, the patient had severe coronary artery disease with a nonrevascularizable main vessel (circumflex artery), restenosis of a secondary vessel, and moderate injury of the anterior descending artery. He was receiving antiplatelet treatment with prasugrel.

Given the restenosis of the secondary vessel, the cardiologist recommended adding ASA 100 mg/24 h to prasugrel before implantation of a drug-eluting stent (the risk of restenosis was high because the vessel was very thin). The patient was referred to our department for evaluation of allergy to ASA and, if the allergy was confirmed, for scheduled desensitization.

The patient reported having been diagnosed with allergy to ASA 20 years previously at another center and that he had experienced recurrent episodes of generalized urticaria with facial angioedema. At that time, he often took ASA as an analgesic and reported that on a few occasions he had experienced lip and eyelid angioedema 1-2 hours after intake. Despite not taking ASA since, episodes of urticaria and angioedema reappeared—albeit less frequently—and resolved spontaneously 7 years ago. He has subsequently tolerated isolated doses of paracetamol, metamizole, and ibuprofen.

We performed a controlled oral provocation test with ASA at 50 mg followed by 100 mg after 2 hours (cumulative dose 150 mg). This dose was initially well tolerated, although 32 hours later the patient presented significant upper lip edema, which resolved spontaneously in 48 hours.

At this point, we considered the possibility that the patient had selective hypersensitivity to salicylates; therefore, together with the cardiology department, we agreed that a good therapeutic option would be antiplatelet treatment with flurbiprofen, an arylpropionic acid derivative that is a potent inhibitor of prostaglandin synthetase and thromboxane A_2. The oral challenge with flurbiprofen programmed for 2 days later (50 mg-100 mg) was positive 10 hours after ingestion of 50 mg the patient developed lower lip edema, which resolved spontaneously in 16 hours.

Hence, the patient was diagnosed with NSAID-induced delayed angioedema. Given the need for antiplatelet therapy with ASA or derivatives, we decided to carry out desensitization to ASA.

Several 2-hour ASA desensitization protocols have been described. Wong et al [5] performed desensitization in 11 patients and Silberman et al [9] in 13 patients. Both authors found their protocols to be successful in most cases, except in patients with chronic idiopathic urticaria. Rossini et al [6] included 26 patients in a desensitization protocol based on doses of ASA similar to those described above, but with an increased interval between the doses. Desensitization was achieved in 5 hours with 88.5% efficiency, although it was unsuccessful in the patients with chronic urticaria and in 1 patient with severe uncontrolled asthma.

We started with a modified version of the protocol of Rossini et al [6] after obtaining the patient's written informed consent. Our modification involved extending the range of doses up to 24 hours and was well tolerated during the first 5 days. On the sixth day, 3 hours after taking 50 mg of aspirin, the patient developed labial angioedema, obliging us to modify our protocol again. Based on the allopurinol desensitization protocol for delayed reactions reported by Fam et al [10], we decided to increase the dose of ASA by 5 mg every 3-5 days. As the patient had experienced a reaction to 50 mg of ASA, we returned to the previously tolerated dose of 37.5 mg for 3 days, with subsequent successive increments of 5 mg. Following this schedule, oral tolerance to ASA 100 mg was achieved with no new adverse reactions 85 days after starting desensitization (Table).

Table. Desensitization Protocol[a]

Day	ASA Dose, mg/d	Tolerance
Day 1	1	Well tolerated
Day 2	3	Well tolerated
Day 3	10	Well tolerated
Day 4	20	Well tolerated
Day 5	37.5	Well tolerated
Day 6	50	Upper lip angioedema
Day 9	37.5	Well tolerated
Day 12	40	Well tolerated
Day 16	45	Well tolerated
Day 19	50	Well tolerated
Day 24	55	Well tolerated
Day 37	60	Well tolerated
Day 42	65	Well tolerated
Day 47	70	Well tolerated
Day 58	75	Well tolerated
Day 63	80	Well tolerated
Day 68	85	Well tolerated
Day 71	90	Well tolerated
Day 75	95	Well tolerated
Day 85	100	Well tolerated

[a]Even though desensitization could have been achieved in 48 days, we had to adjust the protocol according to the patient's availability.

Since achieving tolerance 15 months ago, the patient has been able to take ASA 100 mg/d. A drug-eluting stent was implanted, and the coronary artery disease progressed favorably. Dual antiplatelet therapy with prasugrel was withdrawn after 12 months.

In conclusion, we achieved desensitization to ASA using a customized protocol in a patient with coronary artery disease and NSAID-induced delayed angioedema. However, the underlying mechanism of desensitization remains unknown. We were unable to find similar cases in the literature. The desensitization protocols described usually require adjustments to suit the individual patient's needs.

Funding

The authors declare that no funding was received for the present study.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

Previous Presentation

Data from this study were presented in a brief talk for the Allergy Society of Madrid-Castilla La Mancha interhospital sessions.

References

1. Ayuso P, Blanca-López N, Doña I, Torres MJ, Guéant-Rodríguez RM, Canto G, Sanak M, Mayorga C, Guéant JL, Blanca M and Cornejo-García JA. Advanced phenotyping in hypersensitivity drug reactions to NSAIDs. Clin Exp Allergy. 2013;43:1097-1109.
2. Kowalski ML, Makowska JS, Blanca M, Bavbek S, Bochenek G, Bousquet J, Bousquet P, Celik G, Demoly P, Gomes ER, Niżankowska-Mogilnicka E, Romano A, Sanchez-Borges M, Sanz M, Torres MJ, De Weck A, Szczeklik A and Brockow K. Hypersensitivity to nonsteroidal anti-inflammatory drugs (NSAIDs) – classification, diagnosis and management: review of the EAACI/ENDA and GA2LEN/HANNA. Allergy. 2011;66:818-829.
3. Gollapudi RR, Teirstein PS, Stevenson DD, Simon RA. Aspirin sensitivity, implications for patients with coronary artery disease. JAMA. 2004;292:22-29.
4. Ramanuja S, Breall JA, Kalaria VG. Approach to "aspirin allergy" in cardiovascular patients. Circulation. 2004;110:1524-4539.
5. Wong JT, Nagy CS, Krinzman SJ, MacLean JA, Bloch KJ. Rapid oral challenge-desensitization for patients with aspirin-related urticaria-angioedema. J Allergy Clin Immunol. 2000;105:997-1001.
6. Rossini R, Angiolillo DJ, Musumeci G, Scuri P, Invernizzi P, Bass TA, Mihalcsik L and Gavazzi A. Aspirin desensitization in patients urdergoing percutaneous coronary interventions with stent implantation. AM J Cardiol. 2008;101:786-789.
7. Christou A, Kafkas N, Marinakos A, Katsanos S, Papanikitas K, Patsilinakos S. Rapid desensitization of patients with aspirin allergy who undergo coronary angioplasty. Hellenic J Cardiol. 2011;52:307-310.
8. Lee JKT, Tsui KL, Cheung CY, Chau CH, Chan HL, Wu KL, Cheung GSH, Choi MC, Chan KK, Li SK. Aspirin desensitization for chinese patients with coronary artery disease. HK Med J. 2013;19:207-13.
9. Silberman S, Neukirch-Stoop C, Steg PG. Rapid desensitization procedure for patients with aspirin hypersensitivity urdergoing stenting. AM J Cardiol. 2005;95:509-510.
10. Fam AG, Dunne SM, Lazzetta J, Paton TW. Efficacy and safety of desensitization to allopurinol following cutaneous reactions. Arthritis Rheum. 2001;44:231-238

Manuscript received September 10, 2014; accepted for publication November 24, 2014.

Yosaira Rijo Calderón
Hospital Universitario La Paz
Paseo de la Castellana 261
28046 Madrid, Spain
Email: drarijocalderon@gmail.com

Desensitization to Lenograstim After a Life-threatening Reaction to Filgrastim

Núñez-Acevedo B[1], Rodríguez-Jiménez B[1], Domínguez-Ortega J[2], González-Montellano E[1], Ibáñez-Heras N[3], Enrech-Francés S[4]
[1]*Allergy Unit, Hospital Universitario de Getafe, Getafe, Spain*
[2]*Allergy Department, Hospital Universitario La Paz, Madrid, Spain*
[3]*Pharmacy Department, Hospital Universitario de Getafe, Getafe, Spain*
[4]*Oncology Department, Hospital Universitario de Getafe, Getafe, Spain*

Key words: Lenograstim. Filgrastim. Desensitization.

Palabras clave: Lenograstim. Filgrastim. Desensibilización.

Lenograstim and filgrastim are granulocyte colony-stimulating factors (G-CSF), which are used to stimulate granulocyte production in neutropenic patients, including those with chemotherapy-induced neutropenia. Lenograstim is a Chinese hamster ovary–derived recombinant G-CSF consisting of 174 amino acids with 4% carbohydrate. It is indistinguishable from native G-CSF. Filgrastim is an *Escherichia coli*–derived G-CSF, which differs from lenograstim in that it is nonglycosylated and has an extra methionine group at the N-terminal end of the peptide chain [1].

Although anaphylaxis to G-CSF and granulocyte-monocyte colony-stimulating factor (GM-CSF) has been reported [2-6], desensitization protocols have rarely been proposed [7-8]. We present the case of a 40-year-old woman with infiltrating ductal breast carcinoma treated with total left mastectomy, left axillary lymph node dissection, and chemotherapy (doxorubicin and cyclophosphamide). Filgrastim was given for subcutaneous self-administration at home to treat chemotherapy-induced neutropenia, starting 3 days after the cycle, once a day, for 3-5 consecutive days. Five minutes after the first dose, the patient developed generalized urticaria, abdominal pain, dyspnea, wheeze, hypotension, presyncope, and fecal incontinence. A mobile intensive care unit was called. Profuse sweating, extreme paleness, blood pressure of 80/40 mmHg, heart rate of 54 bpm, baseline oxygen saturation of 95%, and blood sugar concentration of 84 mg/dL were reported. After administration of adrenaline, corticosteroids, antihistamines, fluids, and oxygen, the patient gradually recovered and was taken to the emergency room of our hospital. Temporary leukocytosis with neutrophilia and normal chest x-ray and electrocardiogram findings were recorded. The serum tryptase level was not measured at the time of the reaction. Administration of filgrastim was stopped, and the following 2 cycles of chemotherapy had to be delayed and the dose reduced after the reaction because of neutropenia. Therefore, the indication of G-CSF after the cycle was evident.

A few weeks after the reaction, the patient was referred to our outpatient clinic, where she underwent a complete

Table. Protocol for Desensitization to Lenograstim

Dose	Infusion rate, mL/h	Infusion time, min	Volume, mL	Dose, µg
1	0.5	15	0.125	0.328
2	1	15	0.25	0.65
3	2	15	0.5	1.3
4	4	15	1	2.6
5	8	15	2	5.26
6	16	15	4	10.5
7	32	15	8	21
8	64	15	16	42
9	120	50	100	263

anaphylaxis workup. The findings were within the normal range. The baseline serum tryptase level was 5.7 µg/L. The results of skin prick testing with common aeroallergens (eg, pollen, house dust mites, molds, and animal dander [cat, dog, and hamster]) were negative, and total IgE was 56 kU_A/L. Skin prick testing was performed with commercial preparations (263 µg/mL for lenograstim and 300 µg/mL for filgrastim), as was an intradermal test with concentrations of 1/1000, 1/100, and 1/10 of the commercial preparations, and the results were negative. Given the intensity of the previous reaction to filgrastim and the highly similar chemical structures of lenograstim and filigrastim, a desensitization procedure was carried out with lenograstim (Table). The protocol was designed in cooperation with the pharmacy service. A dose of 263 µg of the drug diluted in 100 mL of saline was used as the infusion solution because this was the maximum dilution that guaranteed drug stability according to the manufacturer's data. The fourth and last cycle of doxorubicin and cyclophosphamide was administered, and lenograstim was planned for 2 days later. The patient was premedicated from 2 days before desensitization with montelukast 10 mg/12 h, acetylsalicylic acid 500 mg/24 h, and ranitidine 150 mg/12 h. One hour before starting the protocol, she received prednisolone 60 mg and dexchlorpheniramine 5 mg. The protocol was well tolerated on the first day of treatment. On the second and third days, the same infusion solution was administered over 50 minutes (120 mL/h), which is slower than normal. On the second day, no reaction was detected, but at the end of the third day, when only a few milliliters of the dilution were left, the patient experienced chest tightness, cough, and shortness of breath that required the infusion to be stopped and salbutamol and adrenaline to be administered. On the fourth and fifth days, lenograstim was administered again following the complete desensitization protocol with no adverse events. The neutrophil count was normal a few days later, and the patient was able to continue chemotherapy with paclitaxel for an additional 12 weeks with good hematologic tolerance.

In summary, we present a desensitization protocol for lenograstim in a patient with a previous life-threatening immediate hypersensitivity reaction to filgrastim. The underlying mechanism of the reaction remains unknown, although it does not seem to be IgE-mediated, considering that it was the patient's first contact with the drug and the result of skin testing was negative. Other mechanisms involved could be nonspecific histamine release, complement activation, and production of cytokines, chemokines, or quinines. Lenograstim is usually administered 2-3 days after the chemotherapy cycle, once daily, and for 3-5 consecutive days. The patient we report tolerated desensitization to lenograstim on the first day with no adverse events. Temporary tolerance to a drug after desensitization can be maintained if the drug is administered at regular intervals, depending on pharmacokinetic parameters and within a time interval of 24-48 hours or 2-3 times the half-life of the drug. Considering that the drug had to be administered daily, we decided to reduce the infusion rate only during the following days of administration, that is, using the same rate as in the last step of the desensitization protocol. The reaction on the third day could have been due to a temporary loss of tolerance that is not easy to explain. Given that the half-life of the drug for intravenous administration is 1-1.5 hours, 24 hours may have been too long to maintain desensitization. In addition, cofactors (eg, infections, nonsteroidal anti-inflammatory drugs, exercise, and stress) could have modified tolerance to previously tolerated doses [9]; however, we were unable to identify the cause in the case we report. The reaction presenting on the third day indicates that a cross-reaction occurred between the 2 drugs and that administration of lenograstim by means of a desensitization protocol was the right option. To our knowledge, this is the first desensitization protocol for lenograstim that has been successful in a patient with a life-threatening reaction to G-CSF.

Funding

The authors declare that no funding was received for the present study.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

References

1. In Hyang Kim, Sung Kyu Park, Ok-Kyung Suh Mi Oh. Comparison of Lenograstim and Filgrastim on haematological effects after autologous peripheral blood stem cell transplantation with high-dose chemotherapy. Curr Med Res Opin. 2003;19:753-9.
2. Tholpady A, Chiosea I, Lyons JJ, Baird K, Leitman SF. Systemic hypersensitivity reaction mimicking anaphylaxis after first filgrastim administration in a healthy donor. Transfusion. 2013;53:1146-7.
3. Batel-Copel L, Mommeja-Marin H, Oudard S, Chauvenet L, Pujade-Lauraine E, Coupier J, Bernadou A. Anaphylactic reaction after a first filgrastim (granulocyte-colony stimulating factor) injection. Eur J Cancer. 1995;31A:2428.
4. Jaiyeslmi I, Giralt S, Wood J. Subcutaneous granulocyte colony stimulating factor and acute anaphylaxis. N Engl J Med. 1991;325:587.

5. Martin-Muñoz R, Gómez-Bellver MJ, Navarro Pulido AM, Orta Cuevas JC. Probable hypersensitivity reaction to filgrastim. Am J Health Syst Pharm. 1996;53:1607.
6. Tulpule S, Shaw BE, Makoni P, Little AM, Madrigal JA, Goldman JM. Severe allergic reaction with anaphylaxis to G-CSF (lenograstrim) in a healthy donor. Bone Marrow Transplant. 2009;44:129-30.
7. Kumar A, Chang T, Chiu A. Desensitization to G-CSF in a patient with clinical hypersensitivity. 2008 Annual Meeting of the American College of Allergy, Asthma and Immunology. Ann Allergy Asthma Immunol. 2009;102:Suppl 1:A30.
8. Shahar E, Krivoy N, Pollack S. Effective acute desensitization for immediate-type hypersensitivity to human granulocyte-monocyte colony stimulating factor. Ann Allergy Asthma Immunol. 1999;83:543-6.
9. Niggemann B, Beyer K. Factors augmenting allergic reactions. Allergy. 2014;69:1582-7.

Manuscript received September 10, 2014; accepted for publication November 24, 2014.

Beatriz Núñez-Acevedo
Allergy Unit, Hospital Universitario de Getafe, Spain
Address: Carretera de Toledo Km 12,5
28905 Getafe, Spain
E-mail: bnunezacevedo@yahoo.es

The New Latex Allergen Hev b 15: IgE-Binding Properties of a Recombinant Serine Protease Inhibitor

Rihs HP, Sander I, Heimann H, Meurer U, Brüning T, Raulf M
IPA - Institute for Prevention and Occupational Medicine of the German Social Accident Insurance, Institute of the Ruhr-University Bochum, Bochum, Germany

Key words: *Hevea brasiliensis*. Hev b 15. IgE-binding. Recombinant allergen. Serine protease inhibitor.

Palabras clave: *Hevea brasiliensis*. Hev b 15. Unión IgE. Alérgeno recombinante. Inhibidor de serina protease.

Serine protease inhibitors (SPI) comprise 60-90 amino acid residues, are frequent in plants, and belong to the plant pathogenesis-related (PR) protein family PR-6. An SPI variant of wheat has been shown to be an important allergen in baker's asthma but not in patients with wheat-induced food allergy [1]. In this study, we elucidate the role of a recombinant SPI variant of *Hevea brasiliensis* in the context of occupational latex allergy. For this purpose cDNA from *H brasiliensis* leaves was used to amplify the SPI-specific sequence. The reaction was carried out in a 50-µL volume containing about 5 ng of *H brasiliensis* cDNA, 5 µL of 10× PCR buffer containing 1.5 mM of $MgCl_2$ (Qiagen), 500 µM of each deoxynucleotide triphosphate (dNTP), 10 pmol of each of the primers Hev b_SPI_SmaI_F (5'-CCCGGGATGGCAAGTCAGTGT-3') and Hev b SPI_SmaI-HindIII_R (5'-CAAGCTTTAGCCAATCRCAGG-3'), and 1.5 units of Taq Polymerase (Qiagen). Reaction products were obtained in a thermal cycler (Life Technologies) with an initial denaturation step (95°C for 5 minutes) and a total of 40 cycles of denaturation (95°C for 1 minute), annealing (56°C for 1 minute), and extension (72°C for 1 minute) followed by 10 minutes at 72°C. The 222-bp PCR product was first subcloned into the pDrive cloning vector (Qiagen). Clones carrying inserts were characterized by restriction analysis and by sequencing on an ABI 310 sequencer (Life Technologies). Sequencing analysis from 4 independent clones revealed that the PCR product corresponded to a 213-bp open reading frame, the sequence of the mature SPI (EMBL accession no. HF937118). The DNA coding for SPI was isolated after separating an Sma I-Hind III digest from the pDrive cloning vector on a 1% agarose gel and subcloned into an Xmn I-Hind III restricted pMALc2 vector (New England Biolabs). The maltose-binding protein (MBP)–rHevb15 fusion protein was expressed in the *Escherichia coli* host NEB5-alpha *F'-I^q* (New England Biolabs) and purified as described for rHev b 1, the recombinant rubber elongation factor of *H brasiliensis* [2], with the exception that a modified column buffer was used (phosphate-buffered saline pH 7.6, 1 mM EDTA pH 8.0). The isolated fusion protein encoded MBP-SPI with an estimated molecular mass of 50.2 kDa (corresponding to 42.7 kDa for the carrier protein MBP and 7.49 kDa for the target protein SPI) derived from the amino acid sequence. The isoelectric point

of SPI was 4.64. A single amino acid residue (glycine) served as a spacer between the MBP carrier and the target *Hevea*-SPI (70 amino acid residues; EMBL accession no. CCW27997.1). Biotinylation of the purified SPI-hybrid and coupling onto streptavidin-ImmunoCAP (Thermo Fisher Scientific) was performed as described with a dilution of biotinylated protein at an optical density of 0.25 at 280 nm [3]. The mean interassay variation of IgE measurements to the MBP-SPI hybrid or to MBP alone repeatedly bound to streptavidin-ImmunoCAPs was 10%. The highest amino acid homology was found in another database entry for an SPI of *H brasiliensis* (GenBank EU295479.1), which was 98% identical owing to a single change in amino acid residue 68 from alanine (GTC) to valine (GCC). In contrast, the SPI of wheat displayed only 36% identical amino acids, and the SPI of castor bean displayed 56% identical amino acids, the highest frequency observed. The total yield of the affinity-purified recombinant MBP-SPI hybrid was 13.9 mg/L according to the results of the Bradford assay with bovine serum albumin as a standard [4].

Serum samples from 21 health care workers with allergic symptoms to natural rubber latex were tested for specific IgE (sIgE) to SPI and 12 available latex allergen components. Values with sIgE >0.35 kU_A/L were considered positive. As shown in the Table, 7 sera (33%) displayed sIgE to SPI (range, 0.56-13.60 kU_A/L). Monosensitization to SPI was not observed. The SPI-sIgE value in relation to the sIgE value of natural rubber latex (k82; Thermo Fisher Scientific) reached a mean percentage of 12% (range, 4%-21%). Detailed analysis of

Table. Characteristics of 7 Health Care Workers With Clinical Symptoms to NRL and sIgE to SPI (rHev b 15)

HCW	Age/ Sex	Total IgE, kU_A/L	Symptoms to NRL	Skin Prick Test to NRL	NRL (k82), kU_A/L	sIgE Reactivity to NRL Allergen Components		
						kU_A/L		SPI[a] (rHev b 15), kU_A/L
#1	40/F	48	U	ND	11.80	nHev b 2 rHev b 6.01 rHev b 1, 3, 5 ,7.02-12	0.66 2.47 <0.35	2.32
#2	20/F	514	A, C, Ez, R, U	ND	92.20	rHev b 1 nHev b 2 rHev b 5 rHev b 6.01 rHev b 7.02 rHev b 3, 8-12	2.29 0.78 22.70 1.81 15.00 <0.35	6.34
#3	44/F	617	A, C, U	Positive	110.00	nHev b 2 rHev b 5 rHev b 6.01 rHev b 1, 3, 7.02-12	7.28 46.40 66.80 <0.35	11.60
#4	25/F	307	A, U	Positive	65.20	rHev b 1 nHev b 2 rHev b 3 rHev b 5 rHev b 6.0 rHev b 7.02 rHev b 11 rHev b 8, 9, 10, 12	1.73 4.84 5.32 8.40 13.90 17.80 1.71 <0.35	13.60
#5	20/F	760	A, C, R, U	Positive	95.10	rHev b 1 nHev b 2 rHev b 5 rHev b 6.01 rHev b 7.02 rHev b 3, 8-12	3.52 18.04 49.60 42.30 3.58 <0.35	7.20
#6	37/F	457	A, C, Ez, R, U	Positive	15.60	nHev b 2 rHev b 5 rHev b 6.01 rHev b 1, 3, 7.02-12	6.26 8.83 4.14 <0.35	0.56
#7	41/F	479	R, U	ND	84.50	rHev b 5 rHev b 6.01 rHev b 7.02 rHev b 1, 3, 8-12	1.86 38.80 12.00 <0.35	7.90

Abbreviations: A, asthma; C, conjunctivitis; Ez, eczema; F, female; HCW, health care worker; MBP, maltose-binding protein; ND, not determined; NRL, natural rubber latex; R, rhinitis; SPI, serine protease inhibitor; U, urticaria.
[a]All MBP-SPI positive sera were tested with MBP and produced negative results (<0.35 kU_A/L).

the 7 SPI-positive sera revealed that all displayed sIgE to rHev b 6.01 (range, 1.81-66.80 kU_A/L) and—with 1 exception—sIgE to rHev b 5 (1.86-49.6 kU_A/L), both of which are known major natural rubber latex allergens in health care workers [5]. Furthermore, sIgE to nHev b 2 (n=6/7) and rHev b 7 (n=4/7) was also frequent. Most notably, the sera with rHev b 7 sIgE showed relatively high values, with a mean of 12.1 kU_A/L and a range of 3.58 kU_A/L to 17.80 kU_A/L. Nevertheless, common sequence features between Hev b 2 and *Hevea*-SPI and between Hev b 7 and *Hevea*-SPI that might explain cross-reactivity or cosensitization were not identified. Control measurements using MBP-ImmunoCAPs were all negative. The results show that the SPI variant studied here is a new natural rubber latex allergen that can now be further tested, for instance, on microarray platforms. In the meantime, the SPI from *H brasiliensis* was reviewed by the allergen nomenclature subcommittee of the World Health Organization/International Union of Immunological Societies and accepted as Hev b 15. In particular, in patients who showed discrepancies between the sum of sIgE values to single natural rubber latex allergens and the IgE values to total natural rubber latex (k82), as shown here for sera #1, 2, 4, and 7, Hev b 15 may be a useful component for improving individual diagnosis.

Funding

The authors declare that no funding was received for the present study.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

Previous Presentation

Preliminary data from this paper were presented as a poster at ISMA, Vienna, December 2013.

References

1. Constantin C, Quirce S, Grote M, Touraev A, Swoboda I, Stoecklinger A, Mari A, Thalhamer J, Heberle-Bors E, Valenta R. Molecular and immunological characterization of a wheat serine proteinase inhibitor as a novel allergen in baker's asthma. J Immunol. 2008;180:7451-60.
2. Rihs HP, Chen Z, Schumacher S, Rozynek P, Cremer R, Lundberg M, Raulf-Heimsoth M, Petersen A, Baur X. Recombinant Hev b 1: large-scale production and immunological characterization. Clin Exp Allergy. 2000;30:1285-92.
3. Sander I, Kespohl S, Merget R, Goldscheid N, Degens PO, Brüning T, Raulf-Heimsoth M. A new method to bind allergens for the measurement of specific IgE antibodies. Int Arch Allergy Immunol. 2005;136:39-44.
4. Bradford M. A rapid and sensitive method for the quantification of microgram quantities of protein utilizing the principle of protein-dye binding. Anal Biochem. 1976;72:248-54.
5. Raulf-Heimsoth M, Rihs HP, Rozynek P, Cremer R, Gaspar A, Pires G, Yeang HY, Arif SA, Hamilton RG, Sander I, Lundberg M, Brüning T. Quantitative analysis of IgE reactivity profiles in patients allergic or sensitized to natural rubber latex. Clin Exp Allergy. 2007;37:1657-67.

Manuscript received November 12, 2014; accepted for publication December 10, 2014.

Hans-Peter Rihs
Institut für Prävention und Arbeitsmedizin der Deutschen Gesetzlichen Unfallversicherung – Institut der Ruhr-Universität Bochum (IPA)
Bürkle-de-la-Camp-Platz 1
44789 Bochum, Germany
E-mail: rihs@ipa-dguv.de